FOREWORD

THIS VOLUME, composed of presentations given by invitation to the symposium on *Viral Encephalitis* at the Fifth Annual Meeting of the Houston Neurological Society, is the third of a series begun in 1955

The organization of the symposium and publication of the material herewith presented could not have been accomplished without the contributions of:

Ayerst Laboratories
Ciba Pharmaceutical Products, Inc
Merck Sharp & Dohme
Pfizer Laboratories
Schering Corporation
G. D Searle & Co
Smith, Kline and French Laboratories
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To them the society expresses its gratitude.

The symposium was moderated by Dr. Russell J Blattner, whose experience and acuity in the field of viral agents contributed significantly to the merit of the discussions.

As with its predecessors, *Hypothalamic-Hypophyseal Inter-relationships* (1955) and *Brain Mechanisms and Drug Action* (1956), this symposium and its published record is testimony to the breadth of view, energy and organizational skills of Dr. William S Fields, who was chiefly responsible for arrangement of the symposium and discharge of the numerous duties incident to its present publication. To him, as well as to the participants, the society expresses its admiration and thanks

GEORGE EHNI, M D
President, Houston
Neurological Society

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INTRODUCTORY REMARKS

RUSSELL J. BLATTNER, M.D.
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DURING the past twenty-five years, outstanding advances have been made in our understanding of the viral group of infectious agents. An important segment of this field of scientific endeavor includes the great volume of highly significant contributions which deal with virus invasion of the central nervous system Nineteen hundred thirty three marked the beginning of this productive investigative era with the definitive isolation of a viral agent from brain tissue recovered from a patient who died with acute encephalitis Since that time numerous fundamental investigations on "encephalitis" have been carried out and recorded (Muckenfuss, R.S , Armstrong, C , and McCordock, H A *Public Health Rep* , U S P H S , *48*:1311, 1933.)

Along with the rapid development of techniques for study of viruses, many aspects of pathogenesis have been clarified, problems of epidemiology solved, etiologic diagnosis of clinical syndromes established, and certain aspects of control, prevention and treatment delineated

This field is an active one and current endeavor in many laboratories promises new contributions It was the purpose of the symposium, which is summarized in this volume, to provide a recapitulation of important aspects of knowledge in this dynamic field, and to supply information concerning recent advances The participants are eminently qualified to do so, and it is hoped that the compilation of the material presented at the Symposium will

prove useful to practicing physicians and other workers in this field.

VIRAL ENCEPHALITIS

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The expression "viral encephalitis," employed in a general sense, means an inflammation of the brain caused by a virus. Often the spinal cord can be involved, either primarily or secondarily, in which case the expression "viral encephalomyelitis" is used These designations are based on clinical observation and, when available, on pathological studies, consequently, they indicate more or less extensive localization of damage in the central nervous system (CNS), with no presupposition as to the virus responsible

Used in this same general sense, the heading "viral encephalitides" can be said to include not only the diseases caused by viruses that are customarily considered to affect primarily the nervous tissue of the CNS, but also those in which the involvement of the brain membranes is perhaps primary, although the nervous tissue is affected as well As an extension of this, it can also include encephalitides that are caused by viruses not ordinarily considered as invading the CNS Finally, certain diseases of the CNS may be mentioned here which are essentially of unknown etiology but, owing to their general aspect, might be of a viral nature

It is not intended to take up all the different diseases that would fall in a chapter of viral encephalitides, as that expression is generally understood, but rather to confine the discussion to a special group of these diseases It might be helpful, however, for a better understanding of the problems involved, to enumerate the more outstanding disease entities that can be or have been included in a general study of the viral encephalitides For the purposes of this presentation, the viral encephalitides can be divided

into two groups: those of known viral etiology, and those in which viral etiology has not been proved. The first group can, in turn, be subdivided. Subdivision (a) includes the arthropod-borne viral encephalitides: Eastern equine encephalitis (EEE), Venezuelan equine encephalitis (VEE), Western equine encephalitis (WEE), Japanese B encephalitis, louping ill, Murray Valley encephalitis (MVE), Russian spring-summer or Far East encephalitis (Russian SS) and St. Louis encephalitis. Several encephalitides (other than spring summer) known to exist in the Soviet Union should perhaps be included here, but information concerning them was not available at the time of writing this article Subdivision (b) consists of encephalitides and other CNS infections caused by viruses not arthropod-borne. In this heterogeneous aggregate can be placed the following diseases or viruses: Coxsackie virus, some of the Echo viruses, encephalomyocarditis, herpes simplex, herpes zoster, infectious mononucleosis (viral?), Lymphocytic choriomeningitis, Lymphogranuloma venereum, measles, mumps, poliomyelitis, rabies and Sabin's B virus Many of these viruses are not considered to be ordinarily encephalitogenic, and some are associated with characteristic clinical entities other than encephalitis

The second main group of viral encephalitides includes diseases for which a viral etiology has, at some time or other, been proposed but that etiology lacks confirmation and is only one of the several advanced for these diseases This is the group of the demyelinating encephalitides, among which are acute primary hemorrhagic encephalitis, multiple sclerosis, postinfection and postvaccination encephalitides

Finally von Economo's disease and Guillain-Barré syndrome can be mentioned, these diseases are neither of proved viral etiology nor of the demyelinating type

Of all these different varieties of encephalitis we would like to discuss the group with which we are most familiar, namely, the arthropod borne viral encephalitides The encephalitic syndrome given by the different viruses in the group is essentially the same with all these viruses when allowance is made for localization and degree of intensity The pathological picture, again in general

terms, is fairly uniform within the group as a whole and is characterized by diffuse rather than localized involvement of brain and cerebellum, the lesions show a degree of neuronotropism but also marked involvement of the supporting elements The meningeal layers present a diffuse cellular infiltration chiefly of lymphocytes and engorgement of blood vessels In the brain tissue itself there is a diffuse infiltration predominantly of the cortical gray matter with lymphocytes and polymorphonuclear leucocytes; perivascular infiltration, small hemorrhages and foci of neuroglial proliferation are conspicuous along with necrosis of neurons and neuronophagia, perivascular demyelination is not seen

Because of this general uniformity, clinical and especially pathological observation can, at best, indicate only that a particular brain infection may be the result of an arthropod-borne virus. The specific diagnosis, if one is possible at all, is to be achieved through laboratory studies, leading either to isolation and identification of the virus or to the detection of antibodies against a given virus in the serum of the patient

The name arthropod borne virus encephalitides was first suggested by Hammon (1943) to describe a number of endemic and epidemic virus infections including EEE, VEE, WEE, Japanese B, louping ill, Russian SS and St Louis encephalitis subsequently, MVE was added to the group Notable advances have been made of recent years in the understanding not only of these diseases and of the viruses that cause them, but also of viruses shown to be related to them but not associated with clinical encephalitis It has, furthermore, become apparent that encephalitis is only one of the forms that infection by the arthropod-borne encephalitis viruses can take, with some of these agents there is evidence that encephalitis is an infrequent occurrence in proportion to the number of persons who have suffered an inapparent infection or perhaps a nonencephalitic type of illness

As an extension of the above concepts, it was not surprising to find that the agents causing arthropod-borne viral encephalitides are, in fact, selected members of a much larger aggregation or family of viruses which we have called arthropod-borne animal viruses (arbor viruses), many of which have no natural capacity

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or tendency to invade the CNS. Under the circumstances, it would seem advisable to broaden the subject of this discussion to include the entire family, rather than restrict it to the more limited encephalitic group.

Arbor viruses are defined as viruses which in nature multiply in the body of arthropods without exerting detectable damage to their tissues or causing other ill effects. It is known for some of these viruses and postulated for others that transmission of the virus to man or other hosts takes place through an arthropod bite, the vector, in turn, becomes infected by ingestion of blood from a host at the time when virus is present in the latter's peripheral circulation.

Antigenic relationships among these viruses, leading eventually to a grouping or classification, have been the subject of study for some time. Thus, cross reactions were shown by Smithburn (1942) between Japanese B, St. Louis and West Nile viruses; by Casals (1943) between louping ill and Russian SS viruses; by Havens and associates (1943) between EEE and WEE viruses; and by Sabin (1948) between some of the viruses causing encephalitides and those of dengue and yellow fever.

A systematic study of the interrelationships among arbor viruses, by Casals and Brown (1954) and Casals (1956), showed that there were at least three sharply defined groups of arbor viruses, designated A, B and C. This conclusion resulted from the study of forty-seven distinct arbor viruses and was based exclusively on the detection of immunological cross reactions. These cross reactions were investigated by different methods, complement fixation (CF), hemagglutination inhibition (HI) and neutralization (NT) tests. It is not intended to describe these methods in detail here, but only to report some of the results obtained.

One fact that soon became apparent, particularly with Groups A and B, was that the HI test showed a wider range of serological overlaps than the CF test, which in turn was more cross reacting than the intracerebral NT test. Hence, the present grouping of the arbor viruses is based essentially on the behavior of these viruses and their hyperimmune sera in the HI test, with the understanding that whatever cross reactions were detected by NT

or CF tests could, in all cases, be demonstrated also by HI tests

Reduced to its essentials, the plan used consisted in preparing by a constant technique hyperimmune sera in animals, usually mice, for each virus under study and testing these sera against as many agglutinating antigens as were available The result of these studies clearly showed, on the basis of inhibition of hemagglutination, that there was a sharp division of the arbor viruses into groups, as illustrated in Table I.

All viruses in Groups A and B developed hemagglutinating (HA) antigens In Group C, however, some viruses were apparently too "weak" to produce utilizable HA antigens, although the immune sera prepared against them reacted well with the available antigens of the same group Failure to produce antigens with the majority of the ungrouped viruses may thus be more a matter of quantity—or technique—than of inherent absence of hemagglutinin; this point is, naturally, of importance and is currently receiving attention Hyperimmune sera against any of the viruses of Group A reacted not only against the homologous antigen but also, to a greater or lesser extent, with all the remaining viruses in the group, while in no instance was a reaction detected against any virus not of Group A A similar thing occurred with Groups B and C Concerning the ungrouped viruses, less work could be done since few antigens are, at present, available; however, none of the hyperimmune sera against these viruses reacted with antigens of either Groups A, B or C

Table I also shows that the encephalitic viruses appear some in Group A—EEE, VEE and WEE, and others in Group B—Japanese B, louping ill, MVE, Russian SS and St Louis. A virus such as WEE, which can cause an encephalitis in man not unlike the one due to St Louis virus, is in its antigenic behavior much closer to Chikungunya virus (which in its known clinical manifestations is similar to dengue) than it is to St Louis virus

The HI reaction with hyperimmune sera has definite advantages for grouping arbor viruses, the cross-reactions detected within Group B, however, are so marked that it is often impossible to arrive at a specific diagnosis with this method In that case, the use of simple immune sera—obtained from experimental ani-

TABLE 1

CLASSIFICATION OF THE ARTHROPOD-BORNE ANIMAL VIRUSES

Group A	*Group B*		*Group C*	*Ungrouped*	
Chikungunya	Bat salivary gl.	SA H 177	BeAn 15	Anopheles A	Eg AR 1152
Eastern EE	Dengue type 1	SA H 336	BeAn 17	BeAn 73	LEN 731-19
Mayaro	Dengue type 2	*St. Louis*	BeAn 848	BeAn 277	Rift Valley Fever
Forest	Ilheus	Uganda S		Be H 151	Sandfly Fever, Naples
	Japanese B	West Nile		Bunyamwera	Sandfly Fever, Sicilian
Venezuelan EE	*Louping ill*	Yellow Fever		Bwamba	SA TAR 53
Western EE	*Murray Valley E*	Zika		California, Hammon Reeves	Tr 3587
	Ntaya			Colorado Tick Fever	TR 8900
	Russian SS			Eg AR 192	TR 9760
	SA TAR 91			Eg AR 1095	Wyeomyia

mals after one injection of antigen or from human cases convalescing from natural infection—may result in a quantitative difference between homologous and cross reactions sufficient to allow a decision Otherwise, an answer may be reached either by CF or NT tests Table II gives results of HI tests with several hyperimmune sera and their corresponding antigens to illustrate the marked relative specificity found in Group A (EEE, WEE) and the lesser degree of specificity seen in Group B

The serological relationships among arbor viruses, which allowed a grouping of these agents, are probably also responsible for the broad serological activity of some human sera thought now to be due to dual infection with members of the same group While it is not possible to describe in detail here all of the observed facts that have been reported elsewhere (Casals, 1957), one in particular might be mentioned It has been shown experimentally—and inferred from studies with field specimens—that inoculation of animals or natural infection of man with two different viruses of the same group at distinct times, can result in an antibody response to all viruses in that group far greater than the sum of the responses given by each virus separately, this pronounced group response is demonstrable by CF, HI and NT tests Even without superinfection, man seems to be sufficiently reactive to infection with one member of Group B to develop antibodies

TABLE II

HEMAGGLUTINATION INHIBITION WITH HYPERIMMUNE MOUSE SERA

Serum	Antigen 8 Units					
	EEE	WEE	Dengue	Japanese	St Louis	Yellow Fever
EEE	*10240	160	0	0	0	0
WEE	80	10240	0	0	0	0
Dengue	0	0	320	80	160	80
Japanese	0	0	320	640	640	160
St Louis	0	0	640	1280	2560	160
Yellow Fever	0	0	40	320	320	320

*Titer of serum = 1 10240
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Semliki Forest	Ilheus	Uganda S		Be H 151	Sandfly Fever, Naples
Sindbis	*Japanese B*	West Nile		Bunyamwera	Sandfly Fever, Sicilian
Venezuelan EE	*Louping Ill*	Yellow Fever		Bwamba	SA TAR 53
Western EE	*Murray Valley E*	Zika		California, Hammon-Reeves	Tr 3587
	Ntaya			Colorado Tick Fever	TR 8900
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	SA TAR 91			Eg AR 1095	Wyeomyia

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*Titer of serum = 1:10240

0 = No reaction at dilution 1:10

Having now reviewed some particular aspects of the work done by numerous investigators in the field of the arthropod-borne viral encephalitides, it may be in order to present the following conclusions:

(1) The arthropod-borne viral encephalitides are caused by a number of viruses which by their behavior in nature and serological properties belong in a larger, more comprehensive assembly or family designated Arthropod-borne animal viruses (arbor viruses).

(2) The arbor viruses can be shown to fall in at least three groups, designated A, B and C

(3) Of the encephalitogenic arbor viruses at present recognized, some belong in Group A. EEE, VEE and WEE; and the remainder in Group B: Japanese B, louping ill, MVE, Russian spring-summer and St. Louis encephalitis viruses

(4) Presumably as a consequence of the serological group relationships, synergistic actions can occur between viruses of the same group when they infect a host As a result, when there is more than one endemic virus from a group in a given area, serious difficulties may arise in trying to interpret, for diagnostic purposes, the results of serological tests This is particularly true with Group B

(5) A survey of the literature shows that inapparent or unrecognizable infection with the agents responsible for the arthropod-borne viral encephalitides is a common occurrence; with some of the viruses, far more frequent than clinical encephalitis

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DISCUSSION

Dr. Blattner, Houston, Texas: It seems to me that this cross-over immunity is a very important practical consideration for us as clinicians, and I would like to know whether the same thing occurs with the inoculation of live virus? I assume that most of this was based on the inoculation or infection with live virus. Now, entering the vaccine era, can you tell us anything about the inactivated virus in cross protection?

Dr. Casals: This was done mostly by injection of live virus, except that hyper-immune sera were actually produced by inoculation first with inactivated material, followed by live virus injection. Our reason for doing this is that mice are susceptible to some of these viruses and we feared that inoculation with live virus would kill them. I would say that I would expect the same thing to occur following injection of inactivated virus.

Dr Haymaker, Washington, D C · Dr Casals concluded that the Guillain-Barré syndrome is a viral disease. I should like to ask Dr Casals to elaborate

Dr. Casals: This classification is presumptive. It is one of these clinical syndromes that is often included in general chapters in which viral encephalitides are discussed I understand that it is more of a radiculitis than an encephalitis, but it is customarily put among these diseases, principally because there is a question of differential diagnosis You have to consider a lot of entities that are not viral in nature, which are often not even encephalitic in nature, to weed them out, so to speak. It is only for that reason that I mentioned the syndrome in this presentation

Dr Imagawa, Houston, Texas· Will you tell us whether there are any tissue cross-reactions as a result of this, and to what extent they may be occasioned?

Dr Casals, No, I don't think that the tissue reaction enters at all in the serological overlap that I have shown here, mainly because one must recognize that everyone of the sera, that have been shown here, have been tested, not only against the antigens of its own group, but against everything else If that were presented in one table it would become really very difficult to present in a

single slide. Every one of those sera (and they were sera obtained in animals inoculated repeatedly with brain tissue infected with a virus) were tested against Group A antigens, against Group B and against Group C, and only the significant part of the test has been presented here. The above is concerned with the hemagglutination inhibition. Concerning the complement fixation test, no serum should ever be tested unless there is a control antigen prepared in exactly the same manner as the active antigen. In the case of a neutralization test, it is possible to have some nonspecific action influencing the result of the test, but one has to have adequate control. So, I believe one can leave out tissue reaction if the tests are well controlled.

ARTHROPOD-BORNE VIRAL ENCEPHALITIDES

Epidemiology of Western Equine and St. Louis Encephalitis

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I have been asked to discuss today some of the major facets of the epidemiology of the viral encephalitides. The qualification "viral" at once denotes that the conditions to be discussed are etiologically restricted to a particular group of biological agents Before further restricting the scope of today's discussion from the standpoint of the etiologic agents to be discussed, I should like to make a few comments germane to the subject of encephalitis as a whole. Etymologically, the term "encephalitis" means inflammation of the brain and nothing more. Consequently, appending this label to a clinical syndrome, or a histopathologic alteration, conveys no information as to the cause which brought about the condition Since encephalitis can be evoked by a number of quite different factors, for example, toxic gases, noxious chemicals, biological agents, it is not always easy in any given case to determine what the underlying cause might be. Consequently, general classifications such as "infectious encephalitis" or 'toxic encephalitis" are frequently used, and while they are preferable to the adjectivally unqualified designation "encephalitis," they do not contribute notably to the precise knowledge of etiology required for the development of specific therapy and for prevention and control of a disease To attain these objectives, it is necessary that specific identification of the etiologic agent be attempted in every case of encephalitis

And now to return to the encephalitides of viral origin While infection with any one of a number of viruses, for example, measles, varicella and influenza may eventuate in an encephalitis, infection with these agents does not generally take this clinical form. the encephalitic end-result in these infections is an incidental event, and hence the respective viruses are not classified as highly, or primarily, encephalitogenic On the other hand, the encephalitogenic capacity of certain viruses appears to be an important basic and inherent property of the agent. Clinically-apparent infection with these agents progresses to a frank encephalitic syndrome in so large a proportion of individuals that the encephalitogenic propensity of these viruses is recognized in the designation 'encephalitis viruses' The most important encephalitogenic viruses are insect-transmitted, so that the diseases they produce are often collectively spoken of as the "arthropod-borne viral encephalitides"

In North America, only three arthropod-borne viruses have a role of any consequence in the production of encephalitis, namely, the Eastern equine encephalomyelitis virus, the Western equine encephalomyelitis virus and the St Louis encephalitis virus While the Eastern equine encephalomyelitis virus has been reported present as far west as Texas the disease caused by this virus will not be considered here because of the surprising paucity of information concerning its epidemiology Attention will be focussed chiefly on Western equine and St Louis encephalitis, since both are diseases of some importance and interest in the western and southwestern United States and both have been studied intensively for some years

HISTORICAL

In 1931, Meyer, Haring and Howitt[1] isolated from the central nervous system of horses with an encephalitic disease a virus which has since come to be known as the Western equine encephalomyelitis virus It was suspected that the virus might be involved in the causation of human as well as equine illness, and this suspicion was confirmed in 1938 when Howitt[2] recovered the virus from the brain of a child

In 1933, an epizootic of encephalitis occurred amongst horses along the eastern seaboard; TenBroeck and Merrill[3] and Giltner and Shahan[4] recovered from the central nervous system of affected animals a virus which was found to be immunologically distinct[3] from the virus reported from the western United States and, consequently, was designated as the Eastern equine encephalomyelitis virus Its relation to human illness was demonstrated by Fothergill *et al.*[5] and by Webster and Wright[6] in 1938, when both groups of workers independently recovered the virus from fatal human cases during the course of a small epidemic in Massachusetts

St. Louis and Kansas City, Missouri, and especially the former city, suffered a large epidemic of encephalitis in 1933. Muckenfuss *et al.*[7] and Webster and Fite[8] recovered a virus from the central nervous system tissues of fatal cases. The recovery of an agent, later proved to be etiologically involved, together with the fact that the clinical picture differed from that of lethargic encephalitis, lead to designation of the disease as St. Louis encephalitis

GEOGRAPHIC DISTRIBUTION

Until about 1939, the Appalachian Mountain range was regarded as a barrier dividing the operational domains of the Eastern and the Western equine encephalomyelitis viruses Over the intervening years, however, the Eastern virus seems to have spread westward, originally believed restricted to the eastern seaboard, it has been detected as far west as Michigan in the north and Louisiana and Texas in the south. The Western equine encephalomyelitis virus has been found, over the same period, to occur in all of the states between the West Coast and the Mississippi River, and east of this river as far as Michigan in the north, and Alabama and Florida in the south In some states, therefore, both viruses are present It should be pointed out that in most of the areas I have mentioned, manifestations of viral activity are confined entirely, or almost entirely, to the occurrence of cases in horses, the *equidae* thus serve as useful sentinels for the detection of virus, at least when it exhibits a high level of activity.

Since the St Louis encephalitis virus, unlike the Western equine virus, does not, in general, tend to produce clinically ap

parent infections in equine species, information as to its geographic distribution is very limited The virus is present along the Pacific Coast, in the southwest and to an unknown extent in the midwest (Missouri and probably some contiguous states)

IMPORTANCE OF THE VIRAL ENCEPHALITIDES

We might at this point attempt a rough assessment of the relative public health importance of the two viral encephalitides with which we are primarily concerned.

In a recent discussion of the encephalitis problem in the United States, Reeves[8] mentions that the attack rate in the encephalitis epidemic in St. Louis in 1933 was 100 per 100,000 population. This attack rate is based not on laboratory-proved cases of St. Louis encephalitis but on the total number of cases of encephalitis *reported* to the health agencies In 1952, a large outbreak of encephalitis occurred in California[10] and the incidence of *reported* cases of encephalitis within the Central Valley, the area of high endemicity of Western equine and St Louis encephalitis, was approximately 42 per 100,000 population Computation of the morbidity on the basis of laboratory-proved cases of St Louis encephalitis gave an attack rate of 2 3 per 100,000 In 1954, the total number of laboratory-proved cases of St Louis encephalitis for California was 99, the attack rate, based on 1952 population estimates was 5 5

As to Western equine encephalitis, a very large epidemic occurred in 1941 in the North Central states and the adjoining provinces of Canada The attack rates ran as high as 22 per 100,000 population[8], which is not too dissimilar from that observed in California in 1952, when the incidence of laboratory-proved cases of Western equine encephalitis in the Central Valley was 20 4 per 100,000 population During "non-epidemic" years, the incidence of Western equine encephalitis in the Central Valley, an endemic area, may be as low as 1 per 100,000 or less From the national standpoint therefore, the arthropod-borne viral encephalitides hardly constitute a public health problem of pressing importance Nor do these encephalitides possess a major significance from the local standpoint either.[11] outbreaks of epi-

demic proportions appear to be an uncommon phenomenon and, in endemic areas of Western equine and St. Louis encephalitis, the activity of the viruses is usually expressed by the occurrence of sporadic cases or, on occasion, the occurrence of what might be regarded as minor outbreaks Both diseases, nevertheless, are greatly feared by the public in those endemic areas where epidemics or outbreaks of some size have occurred, and not only because of the immediate effect on the family, but also because of the possible after-effects of the disease in the victim. I have in mind the mental disturbances and psychiatric problems that may develop as post-encephalitic sequelae,[24] especially in very young children with an encephalitis provoked by the Western equine virus.

THE EPIDEMIOLOGY OF WESTERN EQUINE AND ST. LOUIS ENCEPHALITIS AS OBSERVED IN CALIFORNIA

Although the Western equine and St Louis encephalitis viruses are present over a large expanse of the United States, the epidemiologic information available on these diseases has been derived predominantly from specially-oriented investigations which have been conducted in several selected study areas. Extensive epidemiologic field studies have been conducted over many years by Hammon and Reeves and their associates in one such area, viz., the Kern County area of California, and the California State Department of Public Health has collected certain ancillary data which supplement or complement the data of these workers It is the lack of data from other areas, and not provincialism, that impels me to use the California experience and observations to illustrate the epidemiology of Western equine and St. Louis encephalitis I should also like to point out that it would seem reasonable at this point to continue the presentation with a discussion of the transmission of the disease to man, but I prefer to leave this aspect until the end because of certain comparisons it seems desirable to make I shall, therefore, proceed with a discussion of some of the other facets of the epidemiology of these two diseases as observed in California.

Incidence: The occurrence of cases of encephalitis over the

TABLE I DISTRIBUTION ACCORDING TO ETIOLOGY AND YEAR OF ONSET, OF CASES OF INFECTIOUS ENCEPHALITIS CALIFORNIA, 1945-1955

Year	*Total*	*WEE**	*SLE***	*Etiology Undetermined*
Total 1945–1955	2,722	613	337	1,772
1945	302	26	28	248
1946	160	18	10	132
1947	127	32	6	89
1948	71	–	1	70
1949	80	10	21	49
1950	361	88	69	204
1951	145	22	33	90
1952	813	375	45	393
1953	174	14	22	138
1954	353	22	99	232
1955+	136	6	3	127

*WEE = Western equine encephalitis
**SLE = St Louis encephalitis
+ = Figures for this year are provisional

eleven-year period, 1945 through 1955, is summarized in Table I During this interval, 2,722 cases of acute infectious encephalitis were reported to the California State Department of Public Health This figure does not include cases reported as post-infection or post-immunization encephalitis, nor does it include primary encephalitis due to infection with the viruses of herpes simplex or mumps Clinical specimens for laboratory tests were submitted on a large proportion of these patients with the result that a laboratory diagnosis of Western equine encephalitis was made in 613 individuals and a laboratory diagnosis of St Louis encephalitis was made in 337 individuals In the remaining 1,772 individuals, laboratory tests for etiologic agents were negative or no material was submitted for examination, and the clinical and epidemiologic history gave no clue to the cause of the illness, patients in this category are listed under the heading "etiology undetermined" in Table I It will be noted from Table I that there is a wide variation from year to year in the number of cases of

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1947	127	32	6	89
1948	71	—	1	70
1949	80	10	21	49
1950	361	88	69	204
1951	145	22	33	90
1952	815	373	45	393
1953	174	14	22	138
1954	353	22	99	232
1955+	136	6	3	127

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"infectious encephalitis" reported. The highest incidence was in 1952, when 813 cases were reported; the years 1945, 1950 and 1954 also stand out as years of high incidence. The low points appear to have been the years 1948 and 1949

Considerable variation in the annual incidence of Western equine and St. Louis encephalitis is also apparent for the eleven year span covered by Table I. The highest incidence of Western equine encephalitis was 375 cases (diagnosed by laboratory methods) in 1952. The incidence of this disease declined to very low levels in the succeeding three years, whereas the incidence of St Louis encephalitis reached its maximal level during this interval, viz., 99 cases in 1954. This raises the interesting question as to whether the large outbreak of Western equine encephalitis in 1952 served to produce a herd immunity, thus protecting the population at risk against infection with the Western equine encephalitis virus and whether a similar immunization might not have been brought about by the St Louis encephalitis virus subsequent to the high incidence in 1954 Such an interpretation is not on a sure footing in either instance and is certainly on a much more tenuous basis with respect to the St Louis encephalitis virus, more definite information on herd immunity and the extent to which subclinical immunization occurs will be forthcoming only as additional attack-rate data are obtained over the coming years, or as immunity surveys are conducted to determine the incidence of neutralizing antibody in the population The newer, and simplified, tissue culture techniques should make such surveys feasible on a large scale

Also of interest is the large number of cases that fall into the category "etiology undetermined" Whether this group is largely homogeneous with respect to etiology or whether it represents infections caused by a diversity of agents is unkown This aspect will be considered further in a few moments

Geographic distribution: The occurrence of Western equine and St Louis encephalitis in California apparently is restricted to regions which have high summer temperatures and are under extensive irrigation, i.e., the great Central Valley and several counties, such as Imperial and Riverside, outside it In the occa-

sional instances in which individuals living outside the endemic area acquire the disease, they generally give a history of exposure through travel or sojourn within the area. As described elsewhere,[10] nearly 70% of the infectious encephalitis is reported from the San Joaquin Valley, which is the southern portion of the Central Valley drained by the San Joaquin River, and approximately 15% is reported from the Sacramento Valley, which is the northern portion of the Central Valley drained by the Sacramento River This proportional distribution has been consistent over the years and the greater extent to which the San Joaquin Valley is affected by the encephalitides as compared to the Sacramento Valley is shown by the case rates per 100,000 population for the year 1952 Thus, in the San Joaquin Valley the case rate for Western equine encephalitis was 25; for St Louis encephalitis, 4; and for encephalitis of undetermined etiology, 23, the total rate was thus 52 per 100,000 For the Sacramento Valley, the corresponding rates were 10 for Western equine encephalitis, less than 1 for St Louis encephalitis, and 11 for encephalitis of undetermined etiology, thus giving a total rate of 22 per 100,000 Although this was a year of major prevalence of encephalitis, the rates for the corresponding encephalitis categories outside the Central Valley were less than 1 per 100,000

Seasonal occurrence: The seasonal occurrence of Western equine and St Louis encephalitis, as well as of encephalitis of unknown etiology is illustrated in Table II This table covers the eleven-year period 1945-1955 and the cases are allocated to the month in which the onset of illness occurred It will be noted that the occurrence of Western equine and St Louis encephalitis is confined to the interval June through October, and not beyond the first week of November if we include the single case of St Louis encephalitis which out of some 337 cases, had an onset date after October and specifically within the first week in November Serologic tests for the arthropod borne encephalitis viruses are performed on all patients with CNS disease irrespective of the season of the year but no cases have been encountered with onset dates prior to June nor, with the single exception just mentioned, after October Human infections with the Western

TABLE II

SEASONAL DISTRIBUTION, BY MONTH OF ONSET AND BY CATEGORY, OF INFECTIOUS ENCEPHALITIS
CALIFORNIA, 1945–1955

Disease Category	*Jan*	*Feb*	*March*	*Apr*	*May*	*June*	*July*	*Aug*	*Sept*	*Oct*	*Nov.*	*Dec.*	*Total Cases*
	Number of Cases with Onset in												
Western equine encephalitis						38	291	218	61	5			613
St Louis encephalitis							21	125	112	15	1		337
Encephalitis etiology undetermined	57	59	58	68	68	83	326	456	318	125	67	65	1,745*
Totals	57	59	58	68	68	121	641	799	516	175	68	65	2,695*

*Totals do not include 27 cases "encephalitis etiology undetermined" as date onset not stated

equine encephalitis virus first appear in June, increase in numbers to reach a peak in July, remain at a high level through August and thereafter decline, to disappear by the end of October. Human cases of St. Louis encephalitis, on the other hand, do not appear until July, reach a peak in September and disappear by the end of October. Thus, the peak incidence of the two types of infection is separated by a period of about two months. Apropos this temporal relationship, infection with these viruses is first evidenced in the mosquito population, is subsequently followed by the appearance of equine cases, at least in the instance of the Western equine virus, and human infections follow the appearance of the equine infections. In the mosquito population, as in the human population, infection with the St. Louis encephalitis virus appears somewhat later than infections with the Western equine encephalitis virus.

It is also of interest that encephalitis of unknown etiology has a prevalence pattern indicative of seasonal occurrence. As shown in Table II, the incidence seems to be at a uniform level throughout the year except for the summer and early fall months, at which time there is a very marked increase in the number of cases simultaneously with the increase in cases of Western equine and St. Louis encephalitis. It is not impossible, indeed it is quite probable that some of these cases represent poliomyelitis and especially aseptic meningitis. It is obvious that elucidation of the etiologic agents represented by this catch-all group will require comprehensive laboratory investigation, since the possible role of viruses in the poliomyelitis, Coxsackie and ECHO groups, for example will have to be ascertained and evaluated. Finally, it is not impossible that an as yet unidentified arthropod-borne virus plays a role in this disease complex.

Age and sex distribution: Table III gives the age distribution of cases of Western equine and St. Louis encephalitis. The tabulation gives the age distribution as observed during the major outbreak of encephalitis in 1952 and also summates the age distribution of laboratory-proved cases over the nine-year period 1945-1953. Table III shows that Western equine encephalitis apparently has a predilection for the very young and for those over fifty

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years of age. Thus, of the laboratory-proved cases of Western equine encephalitis, nearly one-half (47.7%) were under the age of ten years, slightly more than one-third were under four years of age and approximately one-fourth (27.7%) were under one year of age. The tendency of the Western equine virus to strike the very young is indicated by the fact that more than two-thirds (69%) of the infant cases were in individuals under three months of age. This is further illustrated by the fact that the attack rate for the Central Valley was 249 per 100,000 population for the age group under one year and 20 per 100,000 for all age groups combined. The accumulated statistics for the nine-year period very

TABLE III. AGE DISTRIBUTION OF CASES OF WESTERN EQUINE AND ST. LOUIS ENCEPHALITIS. CALIFORNIA, 1952 AND 1945–1953

Year and Age	*Western Equine Encephalitis* *Cases*	*Per Cent*	*St. Louis Encephalitis* *Cases*	*Per Cent*
1952 total, all ages	375	100.0	45	100.0
Under 1 year	104	27.7	0	—
1–4	44	11.7	8	17.8
5–9	31	8.3	4	8.9
10–19	36	9.6	4	8.9
20–29	20	5.3	7	15.6
30–39	21	5.6	10	22.2
40–49	27	7.2	5	11.1
50 and over	92	21.5	7	15.6
1945–53 total, all ages	585	100.0	235	100.0
Under 1 year	154	26.3	3	1.3
1–4	74	12.6	38	16.2
5–9	48	8.2	26	11.1
10–19	66	11.3	35	14.9
20–29	31	5.3	37	15.7
30–39	33	5.6	36	15.3
40–49	48	8.2	23	9.8
50 and over	131	22.4	37	15.7

Reproduced from Longshore *et al.*, *Am. J. Hyg.*, 63:69-86, 1956.

closely approximate the data for the single year 1952, as is shown by a comparison of the attack rates presented in Table III.

With respect to the St Louis encephalitis virus, this agent appears to show no striking age selection comparable to that shown by the Western equine encephalitis virus, with the possible exception of the age group under one. It will be noted from Table III that in 1952 no cases of St. Louis encephalitis were encountered in individuals under one year of age, and over the nine-year period 1945-1953, only three cases were found, these three cases represented only slightly more than 1% of the total of 235 cases of the disease encountered during this time interval. The reason for the difference in attack rates of Western equine and St. Louis encephalitis in children under one year of age is unknown One proffered explanation is that subclinical infection with the St. Louis encephalitis virus may occur more widely than does subclinical infection with the Western equine encephalitis virus; consequently, passively acquired immunity in the newborn may be considerably higher to the St. Louis encephalitis virus than to the Western equine encephalitis virus[31]

The incidence of St. Louis encephalitis is approximately twice as high in males as in females, the ratio observed in laboratory-proved cases of this disease over the nine-year period 1945-1953 being 1 8 to 1 This ratio does not seem to be affected by age, whereas the male to female ratio of cases of Western equine encephalitis is markedly influenced by age If all age groups in the 1952 Western equine encephalitis outbreak are considered, the ratio of males to females was 2 to 1. When specific age groups are considered (10), the ratio was 1 to 1 for the age group under four years and became 7 to 1 in the age group five through nine years. The ratio was then approximately 2 to 1 for the age group 10 through 29, jumped to approximately 20 to 1 in the age group 30 to 39, and 26 to 1 in the age group 40 to 49, and finally reverted to 2 to 1 in the age group 50 and over No satisfactory explanation can be advanced for such shifts in attack rates in the male While occupation might conceivably explain the increased attack rate in the 10 to 19 year age group, and probably does account for the very high attack rate in males in the 30 to 49 age group, it can

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The incidence of St Louis encephalitis is approximately twice as high in males as in females, the ratio observed in laboratory-proved cases of this disease over the nine-year period 1945-1953 being 1 8 to 1 This ratio does not seem to be affected by age, whereas the male to female ratio of cases of Western equine encephalitis is markedly influenced by age If all age groups in the 1952 Western equine encephalitis outbreak are considered, the ratio of males to females was 2 to 1 When specific age groups are considered (10), the ratio was 1 to 1 for the age group under four years and became 7 to 1 in the age group five through nine years The ratio was then approximately 2 to 1 for the age group 10 through 29, jumped to approximately 20 to 1 in the age group 30 to 39, and 26 to 1 in the age group 40 to 49, and finally reverted to 2 to 1 in the age group 50 and over No satisfactory explanation can be advanced for such shifts in attack rates in the male While occupation might conceivably explain the increased attack rate in the 10 to 19 year age group, and probably does account for the very high attack rate in males in the 30 to 49 age group, it can

hardly account for the preponderance of males in the five to nine age group.[10] "

Rural and urban factors: Several years ago, we pointed out that employment or residence in rural areas apparently constituted a greater risk to infection than did employment or residence in urban areas. This impression was supported by findings obtained during the 1952 encephalitis outbreak in California A group of 219 cases of infectious encephalitis in Kern County was classified according to etiologic type of encephalitis and by residence of the patient. Classification into rural and urban areas is often arbitrary because it is frequently difficult or impossible to draw distinctions between the two when agricultural pursuits and activities are in intimate relation to individual domiciles and to residential areas. With these difficulties in mind, 82 patients were classified as living in rural of farm areas and 137 were considered to be urban or suburban dwellers This distribution thus gave an overall attack rate for infectious encephalitis of 340 per 100,000 for the rural areas and 61 per 100,000 for urban and suburban areas The higher attack rates encountered in rural areas for "infectious encephalitis" appeared to hold also for Western equine and St Louis encephalitis and for encephalitis of undetermined etiology [10]

The much higher incidence of encephalitis amongst those working or living in rural areas may be due to insufficiency or inadequacy of vector (mosquito) control measures, or to increased opportunities for exposure to the vector in rural as compared with urban areas

METHODS OF TRANSMISSION

Reservoirs of the Virus and Arthropod Vectors

Epidemiologic investigations early suggested that the Eastern and Western equine encephalomyelitis viruses and the St. Louis encephalitis virus were arthropod-transmitted Kelser[11] showed in 1932 that *Aedes aegypti* mosquitoes are able to transmit the Western equine encephalitis virus under laboratory conditions In 1941, Hammon and his co-workers[12] reported the isolation of both Western equine and St Louis encephalitis viruses from naturally-

infected *Culex tarsalis* mosquitoes in the Yakima Valley of Washington. Since that time, investigations of a number of workers, but especially of Hammon and Reeves and their associates, have shown that a number of species of mosquitoes within three genera—*Culex, Aedes, Culiseta*—are capable of acting as vectors of the Western equine and St. Louis encephalitis viruses in the laboratory and that some of these species harbor these viruses under natural conditions. Evidence accumulated over the past fifteen years points to *Culex tarsalis* as the primary vector of both the Western equine and St. Louis encephalitis viruses along the Pacific Coast, and perhaps in other areas where it and the viruses are present. The recent experiments of Barnett[illegible] illustrate the effectiveness of *C. tarsalis* as a vector of Western equine encephalitis virus. His experiments showed that in a very high proportion of instances (107 out of 131 transmissions) the bite from a single infective mosquito would produce viremia and infection in the test host (chick). The minimal extrinsic incubation period was four days, at which time the transmission rate was 10%, and thereafter increased to reach 84% by the thirteenth day. Barnett noted that once the extrinsic incubation period has been passed, *C. tarsalis* tends to transmit the virus as often as it feeds. Since this species apparently remains infective for life, and since it feeds approximately every four days, it is a highly efficient transmitter of infection.

As mentioned earlier, *C. tarsalis* appears to be the primary vector responsible for transmission of the Western equine and St. Louis encephalitis viruses to man and also the equine population. Since these viruses have never been found in overwintering adult mosquitoes, and because there is no evidence for transovarian passage of these agents, other arthropods have been examined with respect to their potentialities as vectors, their ability to harbor the viruses between epidemics or between seasons of activity, or as possible leads to a vertebrate reservoir or reservoirs. These investigations have been directed at various ticks (*Dermacentor andersoni, D. variabilis* and *Argas persicus*), kissing bugs (*Triatoma sanguisuga*) and certain other arthropods.[illegible] The tick, *D. andersoni*, and the kissing bug, *T. sanguisuga*, have been

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DISCUSSION

Dr Kurland, Bethesda, Md.: There are two questions which I wish to raise. First, about 25 per cent of the cases of Western equine encephalomyelitis were said to occur in the group age 50 years and over. At first glance, this appears quite large but then we note this is a relative frequency and not a rate. Might not the percentage reported be due to the large population at risk in the age group 50 years and over rather than any added risk of infection in that age group? Secondly, as the father of four active and inquisitive sons I am impressed by how often they go out to the fields and woods, whereas the girls in the neighborhood tend to stay close to home. I wonder whether the resultant exposure away from home is responsible for the difference in frequency of the illness in the two sexes?

Dr Lennette: Dr Kurland's first question raises an interesting point. Table III gives the age distribution of the cases of Western equine encephalomyelitis in terms of the proportion of the total number of individuals falling within any given age span. Thus the total number of laboratory-proved cases of Western equine encephalitis over 50 years of age was 131 out of the total of 585, or approximately 22% of the total. The lumping of all the age groups of "50 and over" into a single category is a valid procedure since the proportion of individuals falling within the age groups, let us say, from 69 to 79, 79 to 89, etc. is small. Although California, in comparison to many other states, undoubtedly has a larger proportion of individuals in the upper age ranges, I doubt if numerically the group from 50 to 80 years of age, for example, exceeds or even approximates that from 20 to 49 years of age. Off hand, I would say that expression of the occurrence of frank Western equine encephalitis in terms of percentage of cases encountered in any one age group is more meaningful than would be expression of the incidence in any age group in terms of incidence per 100 000 population. It might, of course, be interesting to ascertain the relation of the total number of laboratory-proved cases of Western equine encephalitis in any one age group to the total number of individuals in the population within that

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age range. (The latter information is given in a paper by Longshore *et al.*[30])

The matter of exposure which you raise, Dr. Kurland, represents a very difficult factor to assess. In attempts to evaluate this, as well as other epidemiologic factors, we generally consider the year *1952* by itself and lump together all the data for other years This is because we consider 1952 to be an aberrant year. Normally, *Culex tarsalis* mosquitoes are not encountered in the towns This is because mosquito abatement districts, on one hand, have been organized to combat nuisance mosquitoes and as soon as the citizenry begins to complain unduly about nuisance mosquitoes, control measures are intensified *Culex tarsalis,* on the other hand, generally prefers hosts other than man. In 1952, we had a great deal of rain in the late winter and throughout the spring, which resulted in the flooding of considerable terrain and the mosquito populations became very high In addition, in the southern end of the San Joaquin Valley, two severe earthquakes occurred approximately one month apart Considerable damage to dwellings was done, and fear of additional quakes caused some of the population to sleep outdoors, where they were exposed to attacks by *Culex tarsalis,* which for the first time invaded the towns in quite large numbers This invasion was directly attributable to the heavy population densities in the rural areas and a consequent lack of a normal food supply Even during 1952, however, the ratio of male to female cases was 2 to 1

Dr Blattner It always intrigues me that the migration of birds seems to be associated with encephalitis. Is there any relationship between migration of wild birds and encephalitis outbreaks?

Dr Lennette I do not know of any concrete evidence relating outbreaks of encephalitis to the migration of wild birds The desirability of studying migratory birds as carriers of virus from one area to another has been discussed frequently but so far as I know, no one has actually undertaken such a study I do not mean the intensive studies which have been done on migratory birds in one area or another, but have reference here to studying the birds along the entire migratory flight path in order to determine

where and how the virus is obtained, and how it [illegible]

While the evidence incriminating avian species in the [illegible]emiology of Western equine encephalitis is very [illegible] I think mammals also may be involved as possible reservoirs of the virus. For example, we have just reported the recovery of the Western equine encephalitis virus from five squirrels in California. These squirrels were submitted to the laboratory because they had been observed to behave in an abnormal manner and were suspected of having rabies. Four of the squirrels came from counties where Western equine encephalitis is endemic, but the fifth came from an area where no human cases have been reported. The presence of an infected squirrel in such an area suggests that the virus may be brought in periodically, perhaps by migratory birds and then through the intervention of some arthropod the virus is transmitted from the migratory avian species to the small mammal. This is purely supposition, of course. However we have recently made additional isolations of Western equine encephalitis virus several specimens again having come from non-endemic areas. Recovery of the virus from a small animal such as the squirrel, in a non-endemic area must have some significance. Perhaps the level of viral activity in such areas is not enough to infect mosquitoes and horses or humans. Certainly *Culex tarsalis*, a highly efficient vector, is present.

Dr. Blattner: Did you have any ecto-parasites in that series?

Dr. Lennette: We did not look for ecto-parasites on the squirrels. Actually in most instances we received only the head since it was suspected that the squirrels were rabid and not infected with Western equine encephalitis virus.

the more important ones individually. Some of the more helpful clinical, epidemiologic and laboratory features of the various diseases have been summarized in Table I, along with the recommended specific diagnostic procedures.

I. EPIDEMIC VIRAL INFECTIONS OF THE CENTRAL NERVOUS SYSTEM

A. Poliomyelitis or Enteric Virus Group

1. Poliomyelitis: A great deal has been written concerning the clinical manifestations of poliomyelitis so they will not be discussed in detail here. Infections with the virus of poliomyelitis are common indeed and the majority of the adult population has experienced prior infection with one or more of the three types of poliomyelitis virus, most often without any clinical manifestations of disease Thus, there may be no illness or the patient may complain of no more than a gastrointestinal upset or a mild pharyngitis. If invasion of the central nervous system occurs there is evidence of meningeal irritation with headache, stiff neck and back, and hamstring tightness Paralyses may appear and can result from involvement of any portion of the spinal cord including the medulla and upper brain stem. If the latter portion of the nervous system is invaded regulation of vital functions such as respiration, blood pressure and temperature is often affected It should be pointed out that manifestations of encephalitis, although unusual, do occur in poliomyelitis However, convulsions are most rare unless secondary to anoxia When significant paralysis is evident which is flaccid in type and asymmetric in distribution the diagnosis of poliomyelitis infection may be made with considerable assurance However, if paralysis is absent or slight, the etiologic diagnosis may be difficult Examination of the spinal fluid is the only clinical laboratory test of much value in the diagnosis of poliomyelitis There is an increased number of leukocytes with a total count of from 50 to 100 per cu mm, occasionally higher Early in the disease most of the leukocytes are polymorphonuclear, but within a few days they are replaced by mononuclear cells The protein is moderately elevated with values averaging about 100 mg per cent, and the sugar is normal.

Disease	Clinical Features	Epidemiology	CSF Findings: W.B.C.	CSF Findings: Protein	Specific Diagnostic Procedure: Virus Isolation	Specific Diagnostic Procedure: Serologic
Poliomyelitis	Meningeal signs with occasionally encephalitis. Paralysis common. Convulsions rare	Seasonal in U.S.A. — late summer and fall—may be epidemic	50–100 W.B.C. Predominately PMN early Mono. late	Mod. elevated	Stool* C.N.S. at autopsy	Neutralization* C.F.
Coxsackie virus infection	Meningeal signs sometimes encephalitis. Paralysis rare. May be associated with pleurodynia	Seasonal — summer and fall may be epidemic	Same as poliomyelitis	Same as poliomyelitis	Stool C.N.S.	Neut. and C.F.
ECHO virus infections	Meningeal signs with or without encephalitic manifestations. Some types associated with skin rash. [illegible]	Seasonal — summer and fall may be epidemic	Same as poliomyelitis	Same as poliomyelitis	Stool C.N.S.	Neut. and C.F.
[illegible] encephalitis	[illegible] encephalitis [illegible] more common [illegible]	Mosquito transmitted. Epidemic and sporadic. Common in midwest and western U.S.A.	Similar to poliomyelitis with somewhat higher total count	Same as poliomyelitis	Blood early Brain at autopsy	Neut. and C.F.*
[illegible] encephalitis	Similar to St. Louis	Mosquito transmitted. Birds and horses common animal reservoirs. Epidemic and sporadic [illegible]	Same as St. Louis	Same as poliomyelitis	Blood early Brain at autopsy	Neut. and C.F.*
Rabies	[illegible] Paralysis [illegible]	History of bite of dog or other animal such as fox, cat, skunk, bat	Often normal; may be as many as 100 mono.	Normal or slightly increased	Animal's brain* or saliva. Patient's brain or saliva	
Lymphocytic choriomeningitis	Predominately meningeal. May have symptoms of "grippe"	History of exposure to mice	High: 1000 or more; predominately mono.	Moderately elevated	C.S.F. early	Neut. and C.F.*

tion. Neutralizing antibody to poliomyelitis virus appears early in the course of the clinical illness and thus it is important to obtain the first serum specimen as early as possible. Even so, it is frequently not possible to demonstrate a significant rise in antibody titer. Complement-fixing antibody rises somewhat more slowly, but in some cases never appears in detectable amounts. The results in our laboratory with these two tests in a group of seventy-six patients, all of whom had been shown to have poliomyelitis virus in their feces, indicate that the diagnosis could be confirmed by serologic means in only about one half of the patients and that neither test was significantly superior to the other.

Thus, it would appear that virus isolation is the most satisfactory method presently available for the diagnosis of poliomyelitis infection. Serologic tests are of value, but somewhat less sensitive.

2. **Coxsackie virus infections:** The Coxsackie viruses were first described by Dalldorf and Sickles in 1948. They now make up a group of twenty-four antigenically distinct agents with the property in common of producing disease in the suckling mouse or hamster. They are not infectious for any other experimental animal. On the basis of the lesions produced in the suckling mouse, Dalldorf has further divided them into two groups—A and B. Group A encompasses nineteen types, among which are those viruses that cause herpangina. Group B on the other hand includes only five types and at least three of them have been demonstrated to cause pleurodynia or Bornholm disease.

The suspicion that the Coxsackie viruses could invade the central nervous system of man soon arose since the original isolations were from patients with the clinical diagnosis of poliomyelitis and many of the subsequent isolations were also from patients with non-paralytic poliomyelitis or aseptic meningitis. However, the discovery of a few instances of mixed infections with Coxsackie and poliomyelitis viruses and the demonstration that a certain proportion of the normal population was infected with these agents, made it difficult to relate etiologically the virus in the gastrointestinal tract to the disease of the central nervous system. More recently, Coxsackie viruses have been isolated from

the spinal fluid of patients with aseptic meningitis, which provides conclusive evidence of their causal relationship. To date, Coxsackie A9 and all 5 of the group B agents have been recovered from the spinal fluid The failure to isolate other types of group A viruses may be due in part to the fact that most laboratories conducting such studies employ tissue cultures exclusively and only A9 and the group B agents grow well under these conditions

The clinical features of the central nervous system disease caused by the Coxsackie viruses are not different from those of non-paralytic poliomyelitis Muscular and pleuritic pains are perhaps somewhat more common, and aseptic meningitis may occur in association with the classical features of pleurodynia Some muscular weakness may be noted during the acute disease or early convalescence However, no true paralysis such as is seen in poliomyelitis infection has been described, but further careful follow-up studies are necessary The findings in the spinal fluid are the same as in poliomyelitis

Fatal encephalitis associated with myocarditis in newborn infants due to group B Coxsackie virus has recently been described by Kibrick and Benirschke

The methods for establishing the etiologic diagnosis of Coxsackie virus infections are essentially the same as for poliomyelitis The virus may be present in the nasopharynx, gastrointestinal tract or spinal fluid, but as in poliomyelitis the stool is most likely to be positive. However, if Coxsackie virus is demonstrated in the gastrointestinal tract of a patient, it cannot be accepted with assurance as the cause of the illness for the reasons mentioned above Particularly if any paralysis exists, every effort should be made to determine whether or not there was concurrent infection with poliomyelitis virus Mixed infections of Coxsackie and ECHO viruses also may occur.

Isolation of virus from the spinal fluid is of much more significance etiologically Too little information is available as yet to indicate how frequently this can be done However, it would appear that with present methods virus can be isolated from the spinal fluid in approximately 10% of cases

A rise in serum antibody to the Coxsackie viruses can be

demonstrated during the course of infection by neutralization or complement fixation tests. This is of some value in confirming that a virus isolated did in fact infect the patient, but has the same limitations in diagnosis as stool isolation, since asymptomatic gastrointestinal infections invoke the same antibody response as do those that result in illness. Serologic tests have further practical disadvantages as primary diagnostic methods due to the large number of antigenically distinct Coxsackie viruses.

3. **ECHO virus infections:** The ECHO viruses are a newly recognized group of agents that are widely distributed inhabitants of the gastrointestinal tract of normal people, and are particularly prevalent during the summer and fall They have certain characteristics in common with the Coxsackie viruses, but are not pathogenic for the suckling mouse and can be cultivated only in tissue culture To date, sixteen serologically distinct ECHO viruses have been recognized The letters in the name "ECHO" stand for Enteric, Cytopathogenic, Human Orphans Many of the sixteen types have not been related as yet to any human disease, but a number has been isolated from the spinal fluid of patients with the syndrome of non-paralytic poliomyelitis or aseptic meningitis and some have been demonstrated to be responsible for outbreaks of this disease

Little information is available concerning the clinical manifestations of diseases caused by these agents, but the clinical and laboratory findings do not appear significantly different from those manifested by infection with the Coxsackie viruses

On the basis of observations on patients in our own clinic we have been impressed that those with the aseptic meningitis syndrome from whom ECHO viruses were isolated are more likely to have encephalitic manifestations such as drowsiness, disorientation and convulsions than are those infected with the poliomyelitis or Coxsackie viruses We have also noted edema of the optic nerve in a number of patients in the absence of increased intracranial pressure As with Coxsackie virus infections no severe paralyses have been recognized However, weakness of the muscles of the trunk, shoulder girdle and hip have been observed. The significance of these latter findings can only be determined

TABLE II

THE RESULTS OF VIRUS ISOLATION STUDIES IN PATIENTS WITH ASEPTIC MENINGITIS AT CITY HOSPITAL, CLEVELAND, 1955 AND 1956

Material Studied	No. Patients Examined	No. Patients From Whom Virus Isolated				No. Patients With No Virus Isolated	% Patients Yielding Virus
		Polio	Cox	ECHO	Not Typed		
Stool	196	28 P_1–17 P_2–3 P_3–8	45 A_9–3 B_1–1 B_2–7 B_3–12 B_4–14 B_5–7 Unknown – 1	12 E2–2 E6–1 E7–1 E9–3 E14–5	21	87	56%
CSF	97	0	3 B_4–2 B_5–1	4 E2–1 E9–2 E14–1	2	88	9%

by longer observation of the patients and the accumulation of more control data. Recently there have been several reports, particularly from Europe, of outbreaks of aseptic meningitis in which many of the patients have exhibited a morbilliform rash. The etiologic agent of this disease is a virus classified as ECHO 9, although it apparently has some of the characteristics of a group A Coxsackie virus.

The same difficulties obtain with this group of viruses as with the Coxsackie agents in establishing their etiologic relationship to disease Thus, the isolation of an agent from the feces of the patient does not necessarily mean that it was the cause of his illness. An illustration of the problem encountered in the etiologic diagnosis of patients with the non-paralytic syndrome or aseptic meningitis is presented in Table II, which is a summary of the results of a study conducted at City Hospital, Cleveland over a two year period It is evident that a wide variety of agents, including poliomyelitis, Coxsackie and ECHO viruses has been isolated from the gastrointestinal tracts of approximately 50 to 70% of the patients studied. However, only 9 per cent of the spinal fluids tested yielded a viral agent There is no doubt that Coxsackie and ECHO viruses are capable of producing central nervous system disease in man and probably do so not uncommonly.

B The Arthropod-borne viral encephalitides

St Louis encephalitis, Eastern and Western equine encephalitis, although caused by distinct viral agents, have many features in common. They are transmitted to man by the bite of the mosquito and hence are seasonal in occurrence, appearing during the summer and early fall They are sporadic diseases in certain areas, but may occur in epidemics Although minor differences have been described, there are no distinctive features that would permit clinical differentiation between infections produced by the three viruses Each disease has its characteristic geographic distribution. St Louis encephalitis is largely confined to central and western North America, overlapping with western equine encephalitis which occurs in the Rocky Mountain area and the far West Eastern equine encephalitis, as the name indicates, is large-

ly limited to the eastern portion of the country.

The clinical features of these three types of encephalitis include fever, headache, stiff neck, gastrointestinal symptoms, stupor, convulsions, and coma Various paralyses may be present, usually of the upper motor neuron type. Patients who are more severely ill, regularly demonstrate a significant leukocytosis with a preponderance of polymorphonuclear cells The spinal fluid not infrequently is under increased pressure and an increased number of leukocytes is invariably present. The white cell count in the spinal fluid is usually somewhat higher than that seen in the poliomyelitis group of illnesses, usually exceeding 200 to 300 per cubic millimeter and often rising to levels of 1,000 or more Early in the disease the cells are predominantly polymorphonuclears, shifting to mononuclear cells as the disease progresses The spinal fluid protein content is moderately elevated with levels of approximately 100 mg % The spinal fluid sugar is normal.

From epidemiologic evidence, the etiologic diagnosis may be suspected. However, the clinical features of the disease indicate only the diagnosis of viral encephalitis Special laboratory examinations are required to establish the etiology The only tests of practical value are the serologic ones—neutralization and complement fixation tests Attempts at virus isolation are seldom rewarding, although in rare instances the virus has been demonstrated in the blood and spinal fluid of patients with these diseases In a fatal case the virus may be isolated from the brain by mouse inoculation

C. Von Economo's Disease or Encephalitis Lethargica

Von Economo's disease is an encephalitis, probably of viral etiology, although this has never been established The disease occurred in epidemic form in Europe following the first World War and epidemics then spread throughout the world No epidemics of this disease have been described since 1926, although isolated cases appear that have the clinical features described for this illness The disease was characterized by the symptoms of encephalitis of diffuse nature and ophthalmoplegia was often present Almost any portion of the central nervous system might be involved in this disease, including the spinal cord Sequelae

such as parkinsonism were common. Spinal fluid findings were variable. There was usually a pleocytosis, largely mononuclear cells, accompanied by a slight increase in protein There are no specific diagnostic laboratory tests for this disease. In contrast with the arthropod-borne encephalitides the seasonal distribution was the spring and winter months

II. SPORADIC VIRAL INFECTIONS OF THE CENTRAL NERVOUS SYSTEM

Most of the illnesses listed in this category are relatively uncommon and the pertinent points concerning their diagnoses are indicated in Table I. The discussion is limited to mumps meningoencephalitis since it probably represents one of the most common virus infections of the central nervous system of man.

The true frequency of invasion of the central nervous system during infection with the mumps virus is not known Estimates ranging from 0 5 to 10% have been made, but there seems to be variation with age, sex, season, different outbreaks and other poorly defined factors.

Meningoencephalitis most often occurs two days to ten days after the onset of parotitis, but may be apparent before or at the same time as the parotitis, or it may be the only evidence of infection with the mumps virus

The clinical symptoms of mumps meningoencephalitis are not different from aseptic meningitis or meningoencephalitis due to other causes Headache, vomiting, and fever are the most common Drowsiness, stiffness of the neck, abdominal pain, and irritability are also often present Convulsions are not common, but may occur, particularly in younger children

Examination of the blood is of little diagnostic value. The spinal fluid reveals a pleocytosis of varying degree, but the total cell count is most often in excess of 100 per cubic millimeter Almost without exception the white cells in the spinal fluid are found to be 85% or more mononuclears even early in the course of the disease

The mumps virus may be isolated from the saliva or spinal fluid, but positive results are too infrequent to be of any practical

value. The most useful specific diagnostic tests are the serologic ones. Those most often employed are the complement fixation test and the hemagglutination-inhibition test. The two tests probably detect different antibodies; however, both are usually elevated by one week after onset. As is generally true the demonstration of a rise in antibody level in the patient's serum is the most conclusive indication of infection. The level of complement fixing antibody in a single serum specimen can afford presumptive evidence of recent infection and a serum titer of 1 in 64 or greater may be so interpreted. A refinement of the complement fixation test, developed by the Henles, employs two antigens, a soluble and viral one, and can often provide diagnosis on a single specimen drawn within the first few days of illness. However, this test is not performed in most diagnostic laboratories. The hemagglutination inhibition test is equally as satisfactory. However, results on a single specimen cannot be interpreted due to the high level of non-specific inhibition exhibited by some sera.

III. POST-INFECTIOUS ENCEPHALITIDES

The encephalitides that occur in association with a variety of infectious diseases account for a comparatively large number of the cases of acute disease of the central nervous system in children. The various illnesses with which these are associated include measles, rubella, varicella, vaccination with the virus of vaccinia, infectious mononucleosis, and a variety of others such as influenza, atypical pneumonia, and respiratory infections of unknown etiology. It could be questioned whether or not these diseases should be discussed under the heading of viral infections of the central nervous system since the etiology is not known. Theories concerning their possible etiology are several and include: (1) that the symptoms are due to direct invasion of the central nervous system by the virus which causes the primary illness, (2) that another unrelated agent either coincidently invades the central nervous system, or is activated from the latent state as the result of the systemic infection and (3) that the central nervous system symptoms are the result of an autoimmune mechanism or hypersensitivity to autogenous brain. Most authors seem to favor the last

hypothesis, but it is obviously not within the scope of this presentation to discuss the pros and cons of these various theories.

The symptoms may be primarily meningeal or encephalitic and there may be involvement of the cranial nerves or spinal cord. Convulsions are common in all of the various post-infectious encephalitides

Symptoms of encephalitis of the post-infectious type usually appear from about the third to the eighth day after the onset of the specific illness However, this may vary within wide limits anywhere from preceding the onset of the characteristic eruption in those diseases with exanthemata to as long as twenty-one days after the onset of illness Two of the post-infectious encephalitides might be singled out as varying somewhat from the others in their clinical features Varicella encephalitis in our experience has been associated in the majority of cases with ataxia, often in the absence of any other evidence of cerebellar involvement. This symptom is not common in any of the other diseases of this group The central nervous system complications of infectious mononucleosis are of considerable interest. Meningoencephalitis may occur or the findings may be indistinguishable from the Guillain-Barré syndrome with ascending paralysis, sensory changes and a high spinal fluid protein content without pleocytosis

The frequency with which encephalitis is associated with most of the illnesses mentioned is not accurately known. It is estimated that encephalitis occurs in one out of a thousand patients with measles, but is probably considerably less frequent in the other diseases. Except for infectious mononucleosis mentioned in the foregoing the spinal fluid findings in the post-infectious encephalitides are not characteristic There may be no abnormal findings, but most often there is an increased number of leukocytes ranging from a few cells to a thousand or more The cells may be all mononuclears, or polymorphonuclears may predominate, particularly if the count is high The protein content is usually moderately elevated, and the sugar normal

The diagnosis of post-infectious encephalitis depends largely upon the chronologic association of the skin rash, or other early manifestations of the primary disease and the symptoms of cen

tral nervous system disease. That this is not invariably an accurate criterion is illustrated by the following experience

An infant was brought to the hospital because of a generalized convulsion Two or three days previously the child had developed a vesicular rash characteristic of chickenpox He had not been unusually ill, nor had the temperature been unduly elevated Physical examination revealed no abnormal features except the varicella eruption Lumbar puncture was done and approximately 100 mononuclear cells were present in the spinal fluid and the protein was moderately elevated A tentative diagnosis of varicella encephalitis was made A sibling had developed varicella at about the same time, and although he had not had symptoms of neurologic disease his spinal fluid was examined Much to everyone's surprise this child also had an abnormal spinal fluid with almost identical findings Because of our interest in these children, acute and convalescent serum samples were tested for mumps antibody and clear-cut rises were demonstrated in both patients Only after this was known was the additional information obtained that the children had been exposed to mumps as well as chickenpox about three weeks previously and one of them had had a little swelling of the face a day or two before he was seen at the hospital

This would appear to be an example of a double infection with the viruses of mumps and varicella The meningoencephalitis was most likely due to the mumps virus, but would have been wrongly ascribed to varicella had not the appropriate laboratory studies been done

DIFFERENTIAL DIAGNOSIS

The differential diagnosis of viral infections of the central nervous system from conditions of other etiology may present great difficulty It will obviously not be possible to discuss each of the differential possibilities separately so that we will content ourselves with listing them and little more In Table III this has been done In addition, an effort has been made to indicate those viral diseases that must be thought of if the symptoms are primarily meningitic, encephalitic, or paralytic along with the non viral differential possibilities in each category The list is long, but a number of the non-viral diseases may be quickly eliminated following careful clinical evaluation One of the most important aids in diagnosis is careful examination of the spinal fluid Much confusion can arise if this is not properly done It is not uncom-

TABLE III

DIFFERENTIAL DIAGNOSIS OF VIRUS INFECTIONS OF THE C.N.S.

Etiologic Category	*Meningitis*	*Encephalitis*	*Paralytic*
Viral	Poliomyelitis Coxsackie virus infection ECHO virus infection Mumps Lymphocytic choriomeningitis Herpes zoster Post infectious group ? Herpes simplex	Arthropod borne encephalitides Rabies Herpes simplex Post infectious group Von Economo's Poliomyelitis—occasionally Coxsackie viruses—occasionally ECHO viruses—occasionally	Poliomyelitis Arthropod borne encephalitides Von Economo's Rabies Post infectious group
Non Viral	Treated bacterial meningitis Tbc meningitis Leptospirosis Brain tumors Subarachnoid hemorrhage Multiple sclerosis Torula meningitis Brain abscess Bacterial endocarditis Subdural hematoma Syphilis Osteomyelitis of vertebra Epidural abscess Pneumonia RUL particularly Malaria Trichinosis Rheumatic fever Rheumatoid spondylitis	Bacterial meningitis Tuberculosis Brain tumors Subarachnoid hemorrhage Multiple sclerosis Brain abscess Subdural hematoma Epilepsy Diabetic coma Head trauma Septicemia Degenerative diseases Poisonings Hypoglycemia Acute porphyria	Cord tumor Postictal state Guillain Barré syndrome Post diphtheritic paralyses Herniated intervertebral disc Muscular dystrophies Myasthenia gravis Hysteria Acute porphyria

mon for red blood cells to be mistaken for leukocytes and it is customary in many laboratories to treat the fluid with dilute acetic acid to "lyse" the red cells and make the nuclei of the leukocytes more clearly evident in the counting chamber. If a considerable number of red cells is present it is helpful to centrifuge the fluid and examine the supernatant for xanthochromia in order to distinguish intracranial bleeding from bleeding due to a traumatic procedure. A rapid semi-quantitative determination of the sugar content of the spinal fluid should also be part of the examination It is our practice to use the 5 tube test. This is performed by adding to separate tubes each containing 1 ml. of Benedict's solution 1, 2, 3, 4 and 5 drops of spinal fluid. The materials are mixed and heated in the usual manner. A normal test is one in which some reduction is evident in the 3 or 4 tubes containing the larger amounts of spinal fluid In purulent meningitis or hypoglycemia no reduction will occur in any tube. In diabetic coma, on the other hand, a red precipitate will be seen in all tubes A smear of the spinal fluid sediment should be prepared and stained for bacteria if there is any suspicion of bacterial infection Here, too, care must be exercised in interpreting the findings Inexperienced observers may not recognize bacteria if present or may mistake "dirt" on the slide, precipitated stain and other artifacts for organisms

In inflammatory disease of the central nervous system there is almost invariably an increased number of leukocytes in the spinal fluid There are exceptions, particularly in the post-infectious group, rabies, and possibly a rare poliomyelitis case, although we have never seen a proven symptomatic poliomyelitis infection with normal spinal fluid Certain of the non-viral conditions do not evidence spinal fluid changes These include rheumatic fever, rheumatoid spondylitis, pneumonia, porphyria, trichinosis, the postictal state, myasthenia gravis, the muscular dystrophies, and hysteria Abnormal spinal fluid findings occur in all of the other conditions listed in the table A few deserve special comment

Bacterial meningitis: The usual case of bacterial meningitis with the classical clinical features and a spinal fluid containing many polymorphonuclear cells, bacteria and low or absent sugar

nphasized repeatedly that it is often not possible to make a clin-
al diagnosis more specific than that of viral encephalitis, men-
goencephalitis or meningitis. Since the etiologic diagnosis can
: made as a rule only in retrospect and does not influence man-
;ement of the patient it might be asked whether it is worthwhile
go beyond the clinical diagnosis. There are good reasons why
is desirable to make an etiologic diagnosis whenever possible
hese are.

(1) From the patient's point of view it is often important in
iture management to know that he was previously infected with
particular agent.

(2) From the public health standpoint it is most helpful to
ecognize that a particular disease is occurring in an area This
ay lead to the institution of control measures which will prevent
dditional cases or an outbreak of the disease

(3) Since our knowledge concerning the clinical and epidem-
ologic features of some of the viral diseases of the central nervous
stem is limited, accurate diagnoses are essential to permit the
ccumulation of reliable data

The diagnostic measures available for some of the viral in-
ections have been discussed above and are indicated for others
n Table I Where are such tests performed and how can the aver-
ge physician have them done? In some medical centers research
iboratories will perform a limited number of diagnostic tests
isually in relationship to their research interest Many state
iealth department laboratories do some of the simpler tests for
iral diagnosis and a few such as those in New York and Calif-
ornia have excellent facilities However, even if the state labor-
itory cannot perform the tests the specimen will be forwarded to
he U S Public Health Service facility where they can be
lone The present policy is that all materials should be sent
hrough the state laboratories, and not directly to the Communic-
ible Disease Center or National Institutes of Health

It is of great importance that suitable specimens be obtained
and that they be handled properly For serologic tests, serum spec-
imens must be secured and it is a good general rule that when-
ever materials are submitted for virus isolation acute and con-

valescent sera should also be obtained. The first blood specimen should be drawn as early in the disease as possible and the second approximately one month later. If the materials must be mailed to the laboratory, the sterile serum should be separated from the clot, placed in a sterile test tube and securely stoppered. If the distance is not too great it may be sent by regular mail, but if it is to take much over twenty-four hours it should be refrigerated. If the specimen is not sterile it should be frozen immediately and kept frozen during shipment.

Materials for virus isolation should be properly selected, depending upon the virus that is suspected to be present. It is often well to consult with the laboratory concerning what should be submitted and how it should be sent. Above all, the laboratory should receive adequate clinical information to allow them to perform the appropriate examinations since it is manifestly impossible to test for all of the agents that might cause central nervous system disease. A specimen arriving in the laboratory with the cryptic request "viral studies" is not likely to receive the attention it may deserve.

CONCLUSION

The viruses that can cause disease of the central nervous system are numerous and the differential possibilities even greater. It is the experience of all those interested in this field that even with the most complete study no etiology can be assigned to many of the cases of presumed viral infection. Nonetheless, the etiologic agent can be suspected in many by careful attention to the clinical and epidemiologic evidence and can be confirmed by the intelligent and discriminating use of the virus diagnostic laboratory.

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DISCUSSION OF DR. ROBBINS' PAPER

Dr. Blattner Is there any evidence that there are more than the three types of polio virus?

Dr Robbins Although patients are seen rarely who develop some paralysis following infection with the Coxsackie or ECHO virus, none of these agents could be classed as poliomyelitis viruses The Russians described a type 4 poliomyelitis virus, which I understand has proved to be Coxsackie virus, type A7. To date I think we can safely say that there are only three types of poliomyelitis virus

Dr Florence M Heys, Houston, Texas A question about von Economo's disease Cases are reported from time to time (1951, 1954, 1956) in various parts of the world, which are considered similar to type A encephalitis as it appeared in epidemic form Does this imply that there are endemic foci of the disease, and that epidemics could occur again?

Dr Robbins It is quite true that although no recent epidemics of von Economo's disease have occurred, sporadic cases that resemble it clinically occur throughout the world However, it is not possible to say whether or not this is the same disease since we have no knowledge of the etiology of the epidemic form that occurred in the early twentieth century Those sporadic cases that have been investigated have not yeilded any clues concerning their etiology I suppose that it is perfectly possible that this disease might appear again in epidemic form

Dr Blattner Did you isolate mumps virus from any of your

patients with aseptic meningitis who were studied in 1955 and 1956?

Dr. Robbins: No, we did not, but we made no real effort to do so. The diagnosis of mumps meningoencephalitis was established by serologic tests.

Dr. Heys: Have there been any virus isolations which would support the suggestion that Coxsackie, Type B, might be associated with pericarditis?

Dr. Robbins. The possibility that the Coxsackie viruses might cause pericarditis is a very real one and I suspect that they do on occasions We have not had the opportunity to study any such patients.

Dr. Martha Dukes Yow, Houston, Texas: Have you included tests to diagnose leptospirosis involvement of the central nervous system?

Dr. Robbins: This has not been done on all cases We did, however, obtain agglutination tests for leptospirosis on twelve patients from whom no virus had been isolated. The results were all negative Obviously, this is a possibility that should be explored further, but it did not appear that leptospirosis was a common cause of aseptic meningitis in our area

POSTENCEPHALITIS MANIFESTATIONS OF VIRAL ENCEPHALITIDES*

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SEQUELAE are known to follow virus encephalitides, but reports vary as to their character, incidence, and severity. Most studies of sequelae have been handicapped by small numbers of cases, short periods of time of follow-up, infrequency of observations, or lack of proof of the etiology of the encephalitis. For this discussion of postencephalitis manifestations of virus encephalitis we have reviewed the published reports of several investigators and have added the findings from a study of the sequelae of Western equine and St Louis encephalitis which is currently being carried on in California

The term "virus encephalitis" is most commonly used to denote those encephalitides which are caused by a direct invasion of central nervous system tissue by a virus There may be an encephalitic component in some other virus diseases which are essentially systemic, such as dengue fever, hepatitis, herpes simplex, the exanthemata, and complications of vaccination for smallpox and pertussis In some of these encephalitides an additional biological factor, possibly injury from immunologic developments, may account for the central nervous system complication.

In this paper we shall limit our discussion to the encephalitides in which there is invasion of the cellular elements of the

*Original data were obtained in investigations supported in part by a research grant (B 442) from the National Institute of Neurological Diseases and Blindness National Institutes of Health, U S Public Health Service

central nervous system, particularly neurons, by an identifiable virus.

The three types of known virus encephalitis most commonly recognized in the United States are St. Louis, Eastern equine, and Western equine. During World War II Japanese B also took on significance because it affected troops and civilians working with our troops in the Far East and South Pacific. All of these types occur in epidemics and all are thought or are known to be transmitted by arthropod vectors.

CHARACTERISTIC POSTENCEPHALITIS MANIFESTATIONS

There are differences in the gravity of illness, number of deaths, incidence, and severity of sequelae resulting from infection by these different viruses. The greatest damage seems to be

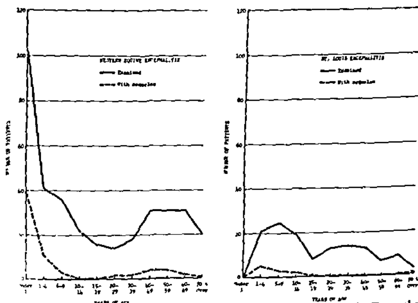


Figure 1 Sequelae of Western equine and St Louis encephalitis From the Encephalitis Clinical Follow-Up Study Stanford University School of Medicine Data based on 18 months of follow-up of patients in California

to the nervous system during its period of very early postnatal maturation Most studies show that the highest frequency and severity of sequelae occur in infant patients, especially those who are ill during the first few months of life (Figure 1). The child, after having reached the age of one year, is much less likely to suffer permanent residuals. Damage is to the motor, emotional, and intellectual spheres Motor sequelae are of the upper motor neuron type, including the pyramidal, extrapyramidal, and cerebellar systems The cranial nerves outside the oculomotor (III, IV and VI) are not frequently affected Convulsions are common sequelae in infant patients (Figure 2). Some investigators have reported a variety of symptom sequelae among older children and

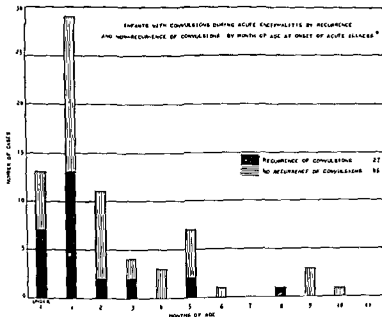


Figure 2 Recurrence of convulsions in infants who had Western equine encephalitis From the Encephalitis Clinical Follow Up Study, Stanford University School of Medicine Data based on 18 months of follow up of patients in California

adults. These include, mainly, headache, sleeplessness, drowsiness, nervousness, depressed feelings, weakness, fatigability, dizziness, irritability, excitability, inability to concentrate, forgetfulness, speech disturbances, difficulty in walking, and tremors These symptom sequelae are probably more often temporary than permanent, lasting usually for a few months to a year or two

STUDIES OF THE SEQUELAE OF VIRAL ENCEPHALITIDES

Research studies of the sequelae of viral encephalitis have varied in their aims, focus, methods, and completeness. Some of the larger studies have included all patients in whom a diagnosis of encephalitis has been made epidemiologically and clinically although confirmation of the etiology in each case has been lacking This might mean that fallacious conclusions have been drawn because it has now been shown that encephalitides caused by at least two known viruses and an undetermined number of unknown viruses may occur in the same epidemic.

Studies which have been limited to cases in which the etiology was proved in each case, have, for the most part, been handicapped because there have been too few patients available for follow-up, or, for one reason or another, patients have not been followed over a sufficiently long period of time

Despite these limitations, studies which have been done have provided some information about postencephalitis manifestations and serve as guideposts for current and future investigators

It was during the encephalitis epidemic of 1933 in Missouri and Kansas that the St Louis virus was identified. In St. Louis, over 1,100 cases were reported during this epidemic. During the Fall of 1934 a follow-up questionnaire was mailed to 874 patients[1] Of the 331 patients who responded, 141 said they felt as well as before the attack and seventy-nine reported their health improved Only twenty-two patients, or 7 per cent, felt physically unable to resume their previous occupations and six of these were over sixty years of age The investigators believed that the infirmities of age may have been as important as the residuals of encephalitis in rendering these patients physically unable to work. Thirty two patients, or 11 per cent of those who answered the

questionnaire, complained of muscular tremors Based on the information given by patients in answering the questionnaires, the authors concluded that the muscular tremors did not suggest parkinsonism

A group of thirty-one patients were later examined by physicians and a number of organic difficulties were uncovered These included speech defect, visual disturbances, difficulty in walking, double vision, nystagmus, paralysis of one or several extremities, hemiplegia, convulsions, and altered reflexes The authors did not relate these sequelae to age On the basis of the data secured on the 331 patients who answered questionnaires, including the thirty-one who were examined by physicians, the investigators concluded that organic residuals occurred in approximately 6 per cent of these patients and none were of great severity Subjective nervous complaints of slight or moderate degree, particularly headaches, irritability, loss of memory, and drowsiness, were fairly frequent residuals Difficulty in walking, which was seldom sufficient to incapacitate the patient, was the most important disturbance of the motor nervous mechanism

Eastern equine encephalitis is usually considered to be the most severe and to have the highest mortality of the known virus encephalitides encountered in *the United States Eastern equine* more frequently attacks infants and children than older persons and is similar in this respect to Western equine. St Louis infection, in our experience, seldom occurs in infants

A study of sequelae was made following the epidemic of Eastern equine encephalitis which occurred in Massachusetts in 1938 There were forty-four suspected cases, but, following investigation, the diagnosis of Eastern equine encephalitis seemed unwarranted in ten cases This left thirty four cases, nineteen of which were confirmed as being caused by the Eastern equine encephalitis virus Fifteen others were diagnosed on the basis of the clinical picture or from the character of the central nervous system lesions About 70 per cent of the patients were children under ten years of age The mortality for the thirty-four patients was 74 per cent The nine surviving patients were followed up over a period of nine years Two died and the cause of death was attributable

directly to sequelae of encephalitis. Three had hemiplegia and mental deficiency or deterioration with two requiring permanent hospitalization. One had chronic epilepsy with mental deficiency and hysterical behavior and in one there were neurologic signs with some emotional instability. Only one of the nine surviving patients was known to have recovered. The ninth patient had not been located.[22]

There have been small outbreaks of Eastern equine encephalitis in the coastal regions of Texas and Louisiana since the Massachusetts epidemic of 1938. In 1955, four cases occurred in Massachusetts which have been reported in the literature. Two of these patients were infants; one died and the other was left with severe brain damage.[4] The other two, who were adults, recovered without sequelae.[5]

One of the earliest reports on sequelae of Western equine encephalitis was in a paper by Eklund, who investigated an outbreak in Minnesota in 1941. He discussed the clinical findings on 314 cases, and stated that sequelae in adults had not been definitely established although one instance of marked personality change had been observed. Mental retardation, convulsions, and spastic paralysis had been observed in four infants. One five year old child developed quadriplegia and sensory loss.[6]

Fulton and Burton,[7] investigating outbreaks of Western equine encephalitis in Saskatchewan, followed up 101 cases of laboratory confirmed encephalitis which occurred between the years 1940 and 1952 and found that fifteen had sequelae. Eight of the fifteen were referred to as children but the age at onset of the encephalitis was not given. These children were diagnosed as having cerebral palsy, of being mentally retarded, and having epileptic seizures. One adult was said to have mental deterioration, one was under treatment for schizophrenia, two had a suggestion of parkinsonism, one had an anxiety neurosis and another a mild personality change. These authors believed that in children the mental and motor changes show up soon after the illness while mental impairment in adults may develop gradually over a period of a year or more.

A study of the sequelae of Western equine encephalitis was

made by investigators from the Departments of Neurology and Psychiatry at the University of Colorado Medical School.[a] Fifteen patients who were ill in August and September, 1949, were observed during the acute phase of their illnesses and were followed for six to eight months in an attempt to evaluate the sequelae of their infections. This series included three infants aged ten weeks and twelve adults aged seventeen to fifty-eight years.

Soon after discharge from the hospital following the acute illnesses, the patients returned for psychologic testing and for one or more neurologic examinations. Six months later they were again given approximately the same battery of psychologic tests, each was interviewed by a psychiatrist, and was examined neurologically. Finally, the relative with whom the patient was living was interviewed. During the interview an attempt was made to evaluate the patient's pre-encephalitic and his postencephalitic adjustment.

All three infants had high fever and focal convulsions during their illnesses. Follow-up studies revealed alert, healthy appearing infants. The principal defect was an inability to use one side of the body as well as the other. One of the children continued to have focal convulsions two to three times a week in spite of adequate anticonvulsant therapy.

Nine of the twelve adults had sequelae; in six these were regarded as severe and in three they were described as moderate. In the other three patients, sequelae were minimal or nonexistent. The severity of sequelae was directly related to the duration of coma experienced during the acute illness. Sequelae consisted of muscle rigidity, speech difficulty, facial masking, loss of weight, and organic impairment of psychologic functioning. The sequelae did not improve and in many instances had progressed. The investigators suggested that the virulence of the virus of Western equine encephalitis may vary with epidemics and an increase in the virulence of the virus in this particular epidemic may have accounted for the severe sequelae observed. Findings from our California study indicate there may be considerable improvement over a period of a year or two in the early benign psychiatric residuals which occur in adults. In view of this, one wonders whether

improvement might have been noted in these adults had they been followed for longer than six to eight months.

A study made of Japanese B encephalitis following the Okinawan epidemic of 1945[7] showed a case fatality of approximately 25 per cent among the 102 native civilian patients who were definitely diagnosed. About a year later thirty-eight survivors were examined for clinical evidence of residual damage to the central nervous system. The investigators believed that their sample of the surviving patients was not large enough to give conclusive information regarding sequelae, but they felt they could safely say that sequelae occurred in not less than 14 per cent. This incidence was higher than that reported by Japanese workers following the larger epidemics of 1924 and 1938. Eleven of the thirty-eight survivors who were examined showed neurologic sequelae. Ten of the eleven patients fell within the age group ranging from four to fifteen years. Three children were severely incapacitated, having marked motor as well as behavior and intellectual impairment. One of these children had convulsions during the encephalitis attack but no mention is made of any recurrence after the acute symptoms subsided. The remaining seven patients had difficulties in motor function such as weakness of one or more extremities and incoordination. Two patients had aphasia and one was described as having a mild personality change. All patients with minor residuals gave histories of gradual improvement and return of function over a period of several weeks to a few months.

THE CALIFORNIA STUDY*

We are now in the fourth year of our follow-up study of the 634 persons who had Western equine and the 350 persons who had St. Louis encephalitis during the past twelve years in Cali-

* The investigators for the "Encephalitis Clinical Follow-Up Study" are Knox H. Finley, M.D., Robert E. Cook, M.D., John M. Harter, M.D., Richard J. Palmer, M.D., and Naomi Riggs, M.S., all from the Stanford University School of Medicine. Assistance is given by Arthur C. Hollister, Jr., M.D., Chief, and William Allen Longshore, Jr., M.D., Assistant Chief, Bureau of Acute Communicable Diseases, California State Department of Public Health and by the staffs of twelve local California Health Departments. The virus studies are carried out in the Viral and Rickettsial Disease Laboratory, California State Department of Public Health, of which Edwin H. Lennette, M.D., is Chief.

fornia Approximately 60 per cent of these patients have been examined in special follow-up clinics—some as many as five or six times at semi-annual or annual intervals. In addition, approximately 150 persons have been examined who had a clinical diagnosis of encephalitis but for whom laboratory tests for etiology were inconclusive.



Figure 3 Location of encephalitis follow up clinics in the California Central Valley

TABLE II

RECURRENCE OF CONVULSIONS IN PATIENTS WHO HAD CONVULSIONS DURING ACUTE ATTACK OF WESTERN EQUINE ENCEPHALITIS BY AGE AT ONSET OF ILLNESS *

Age at Onset	*Number of Patients Examined*	*Number Who Had Convulsions During Acute Illness*	*Number Who Have Had Recurrence of Convulsions #*
Less than 1 year	115	101	35
1 – 4 years	74	30	7
5 – 9 years	62	14	3

* None of the patients who were free from convulsions during the acute illness have had convulsions since

Interval between acute illness and recurrence of convulsions ranged from a few days to two years, except in one case in which the interval was three years and another in which the interval was five years

Encephalitis Clinical Follow Up Study,
Stanford University School of Medicine,

March 1, 1957

ing the acute illness There were eleven patients in this group who did not have convulsions during the acute illness and have had no convulsions since. Of the children whose age at onset was one through four years, seventy-four have been examined thirty had convulsions during the acute illness and seven have had recurrence. Again, of those who did not have convulsions during the acute illness, none have had convulsions since. Of the children aged five through nine years, sixty-two have been examined, fourteen of these had convulsions during the acute illness and three have had a recurrence. In this group also, of those who were free from convulsions during the acute illness, none have had convulsions since. Generally speaking, therefore, it can be said that convulsions have recurred in less than one-third of patients who had convulsions during the acute illness Futhermore, it seems safe to state that regardless of age, convulsions are unlikely to appear as sequelae unless convulsions occurred during the acute illness (Table II).

There is evidence from our study that sequelae among the St Louis patients are less frequent and less severe than among patients who had Western equine encephalitis" This is a weighted finding, however, because all the patients under one year of age, in whom the highest incidence and greatest severity of sequelae occur, had Western equine infections. In California, no more than 10 infants have ever been known to have St Louis encephalitis (Figure I).

The following case descriptions illustrate some of the characteristic postencephalitis manifestations

Case I. (J R.)

A sixty-nine year old man has by far the most severe neurologic and psychiatric impairment of any of the adults in our study series

The history dates back to July 21, 1950 when he felt "poorly" He had aching in the eyeballs, a generalized headache, aching of the joints, and a temperature of 101° F By the following day his fever reached 104° F with still no localizing symptoms and no abnormal physical findings. By the third day his temperature spiked to 105° F and he became incoherent and disoriented He was hos-

The complement fixation titer for Western equine antibodies rose from 1:8 to 1:64 over a period of six weeks. The patient was comatose for eight days. There were tremors of his extremities during the acute illness but these disappeared. A month or two following the illness he developed some weakness of the entire left arm without sensory disturbance.

Upon examination about nine months after his illness, a fine tremor, particularly of the fourth finger of the left hand, was noted. There was no rigidity.

Two years later he had no complaints. However, examination revealed absence of convergence of the eyes and related absence of reaction of the pupils to accomodation. He had a slight but definite impairment of swing of the left arm as compared to the right. At times the left hand, sometimes just an individual finger and sometimes the whole hand, showed an unmistakable rhythmical parkinsonian type tremor. There was a distinct cogwheel phenomenon in the left biceps. In all probability these findings were of an encephalitis etiology. Unfortunately, we will be un-



Figure 5. Case 4 (P.M.) Present age, 11 years. Age 4½ years at onset of Western equine encephalitis. Critically ill with fever up to 105°F and stupor for two to three weeks. Pyramidal and extrapyramid [illegible] duals but no apparent behavior retard[illegible] [illegible]lectual [illegible] The athetoid hyperflexion of the right fin[illegible] [illegible]d elbo[illegible] Also, note the weight bearing on the r[illegible] [illegible]and [illegible] ace-ment of the right leg and eversion [illegible] [illegible]le du[illegible]

able to follow this patient as he died in August, 1955, following a cerebral vascular accident.

Case 4. (P. M.)

A four and one-half year old girl who had Western equine encephalitis in July, 1950 had a severe illness with fever up to 105° F A lumbar puncture was done five days following the onset of illness and the spinal fluid showed 98 WBC, 26 per cent being polymorphonuclears There was a rise in complement fixing antibody titer for Western equine virus between the acute and convalescent blood specimens from less than 1:8 to 1:512. The patient was hospitalized for almost three weeks and during most of this time was comatose and stuporous. She did not have convulsions but upon regaining consciousness it was apparent that she had an aphasia as well as a right hemiparesis

Upon returning home from the hospital she was irritable, childish in her behavior, and still had marked speech impairment Gradually her speech returned and three years following her illness she seemed to be bright, alert, attentive, and cooperative, however, she had a marked athetoid movement of the right arm as well as athetosis of the right leg associated with some spasticity of the right leg

She has been seen periodically in our special encephalitis clinics—the last time when she was almost eleven years old or six and one half years after her encephalitis She has been making good progress in school and has adjusted very well to her remaining athetoid difficulty.

Case 5. (M. P.)

A boy of two and one-half months became ill on July 19, 1952 and was admitted to the hospital two days later His fever was 105° F and remained at a high level for six days He had frequent convulsions of short duration for four or five days. During these convulsions his head turned to the right, his eyes rolled back, and the whole right side of his body was more involved than the left The convulsions were associated with vomiting He also was lethargic, irritable, and had generalized rigidity

The spinal fluid on the day of admission to the hospital contained 1,080 WBC, 85 per cent of which were polymorphonuclears Sugar was 74 mg % and quantitative protein was 58 mg %

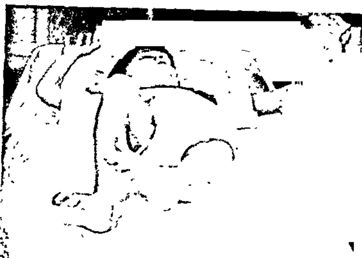


Figure 6 *Case 5* (M P) Present age, 4½ years Age 2½ months at onset of Western equine encephalitis Critically ill with fever spiking to 105°F. for 6 days Frequent convulsions He is now a small, immature, poorly developed child Unable to sit unsupported Marked intellectual impairment Spasticity and rigidity of all four extremities with very little voluntary control Both pyramidal and extrapyramidal tract signs of all extremities

Ten days later a second spinal tap was done and the fluid contained 80 WBC, 68 per cent of which were polymorphonuclears Blood was not drawn for virus study until a month after the onset of the illness, at which time the complement fixing titer for Western equine virus was 1 256 or more Five months later the titer was 1 512 Blood drawn a year later still maintained a complement fixing antibody titer of 1·256

This child has been seen at regular intervals since his illness, the last examination being made when he was four and one-half years old He is a very small, immature, poorly developed child His head is slightly asymmetrical He is extremely spastic in all four extremities and in the trunk Any slight movement in the environment causes him to produce a masslike extension reflex. He is unable to sit unsupported and spends most of his time on his back He makes very little noise and drools a good deal He has no apparent seizures He has made little, if any, progress in mental or motor development. He is still being cared for at home but eventually will require institutional care.

Cases 6 and 7. (R. C. and T. C.) (See Table I)

Fraternal twin girls who were born on August 2, 1952 became ill at five days of age. Both were acutely ill with high fever, lethargy, neck rigidity, dehydration, and bulging fontanels.

Lumbar puncture on one child showed a spinal fluid with 695 WBC, of which 58 per cent were polymorphonuclears. Sugar was 61 mg % and protein was 210 mg %. The spinal fluid on the other twin showed 511 WBC, of which 76 per cent were polymorphonuclears. Sugar was 68 mg % and protein was 238 mg %.

The hospital course for both infants was stormy. Temperatures ran from 101° F. to 104° F. for three days. They suffered repeated convulsive episodes. They were extremely lethargic and refused formula. Clinical improvement was noted on the seventh day and they were discharged, apparently well, after thirteen days hospitalization.

There was a rise in complement fixing antibody titer for Western equine virus on blood specimens taken during the acute and convalescent phase of the illness in one child from less than 1:8 to 1:128 and then to 1:512, and in the other child from less than 1:8 to 1:16 to 1:256. Blood drawn on the mother showed complement fixing antibody in a titer of 1:128. Without doubt this was a case of transplacental transmission of the virus from the mother to the twin infants.[13]

These children have been examined at intervals since their acute illness and were last seen at the age of 4 years. Both children are extremely restless and have definite neurologic and behavior residuals. Although both children are walking, they are quite unsteady. One does not talk at all and the other says only a few words. One is able to partially feed herself, but the other, who is more severely damaged, is unable to do so because of the spastic right side. Neither child has acquired toilet habits. Both continue to have convulsions periodically.

SUMMARY

All the known virus encephalitides appear to leave sequelae in the motor, intellectual, and emotional spheres. There is considerable variation in the incidence and severity of the sequelae. While the difference in virulence of the different viruses, or the difference in virulence of the same virus in different epidemics,

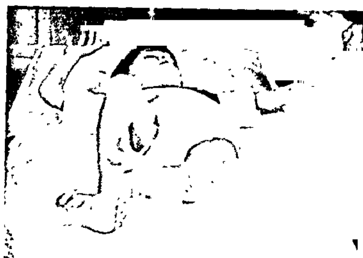


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SUMMARY

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undoubtedly is a factor in accounting for variations in sequelae, the factor which is most impressive is the age of the patient at onset of the encephalitis. Both Eastern and Western equine encephalitis frequently lead to severe sequelae in infant patients For example, in the California study over 50 per cent of infant patients—those less than one year of age at onset—who had Western equine encephalitis have been found to have severe sequelae Japanese B encephalitis also seems to be more damaging to children than to adults. Reports of sequelae, following Eastern equine in adults are too limited to be conclusive, but Western equine does not appear to leave severe sequelae in older children and adults. Again reporting from the California study, in older children—those between one year of age and adolescence—sequelae from Western equine infection occur in not more than 20 per cent and the residuals are of slight to moderate degree. Sequelae occur in less than 20 per cent of adults and in most patients the residual effects are of such slight degree that they are of no practical significance to the patient.

Follow-up of St Louis encephalitis patients indicates that sequelae are infrequent and none are of great severity In California, the St Louis virus only rarely affects infants, which may in part account for the lower rates of residuals as compared with Eastern and Western equine encephalitis

Of especial interest is the occurrence and recurrence of convulsions In our California study, convulsions during the acute illness occurred with decreasing frequency as age increased—in 90 per cent of patients under one year of age, in 41 per cent of patients one through four years, in 23 per cent of the five through nine year olds, and rarely in adults Recurrences also occur in decreasing proportions It is not known how long a lapse may occur between the acute illness and the onset of convulsions as a sequela. It will be of interest to follow patients who were ill in infancy through their adolescent years to learn if convulsions appear. It seems safe to say that regardless of age, convulsions are unlikely to appear as sequelae unless convulsions occurred during the acute illness

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DISCUSSION OF DR. FINLEY'S PAPER

Dr Earle, Galveston, Texas: Have any of your children shown any progressive advancing picture, and do you believe there is any such thing as chronic viral encephalitis?

Dr Finley None of our children have shown a chronic progressive picture I pointed out that some of the children, particularly infants, will not appear to be too severely handicapped when they leave the hospital, but later will I believe it is not so much a problem of progression, as it is our inability to observe the results of the CNS damage until the child is older and we find that he is unable to cope with the increasing demands in his en-

vironment. There may be chronic progressive viral encephalitis but we have not seen this pattern in the encephalitis caused by the Western equine or St. Louis viruses.

Dr. Fields, Houston, Texas: In Guam, during December 1956, I saw about a half dozen patients who had had encephalitis during the epidemic of 1947/48, both Japanese B and mumps varieties, some with both infections simultaneously. The presumption is that those who had encephalitic sequelae had them as a result of the Japanese B rather than the mumps invasion of the nervous system. There is a survey underway now to determine what percentage of these patients did suffer sequelae in the long term. Some have died; two or three who died were subsequently reported to have developed parkinsonism during the period between 1918 and 1956

Dr Jones, St Louis, Missouri: I had the experience of following numerous patients during and after the epidemic of lethargic encephalitis which occurred in this country between 1919 and 1925 Many of these patients exhibited a variety of neurologic states—so-called residuals These residuals were marked from time to time by exacerbations and changing symptomatology. In our experience in the St Louis area, the most common chronic residual was the Parkinson syndrome In addition, we saw other forms of spontaneous movements and oculogyric crises

The "organic" mental syndrome so common in lethargic or winter encephalitis, was noted infrequently in the St Louis encephalitis epidemics of 1933 and 1937 Our experience has also indicated that the so-called residuals of St Louis encephalitis are inconsequential in comparison with those seen in the lethargic type I do not know of a single case of parkinsonism having been reported after either the 1933 or 1937 epidemics in St Louis (See Jones, A B and Bozalis, G S 1937 St Louis Epidemic of Encephalitis *J Missouri State Med Assn* pp 5-7, January, 1940)

Dr Finley: We have observed about 200 adults for four years and some fifty or so others at intervals fro to twelve years following their encephalitis attacks, and possible exceptions we have obse arkinsonist

PATHOLOGY OF VIRAL DISEASE IN MAN CHARACTERIZED BY NUCLEAR INCLUSIONS

With Emphasis on Herpes Simplex and Subacute Inclusion Encephalitis

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AND

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THIS presentation is concerned with viral diseases characterized by the presence of nuclear inclusions, but emphasis is given those in which the central nervous system is affected Certain pulmonary infections are dealt with because the cases reported have been too few to permit the neurotropic potentialities of their causative agents to be ruled out Salivary gland virus disease is included for purposes of differential diagnosis

The subdivision of viral nuclear inclusions into Types A and B by Cowdry[n] still serves a useful purpose Type A *homogeneous* inclusions usually fill the nucleus, though a narrow clear space, or halo, may be seen between inclusion and nuclear membrane The nucleolus is usually displaced to the periphery of the nucleus In the brain the inclusions are either eosinophilic or amphophilic, with a dull red cast, while in other organs they are usually eosinophilic The nuclear membrane is thickened as the result of the accumulation of basichromatin (basophilic chromatin), often in

Figure 1 *A* (Case 5, Table III) Type A homogeneous nuclear inclusion with halo, and spherical cytoplasmic inclusions *B and C* (Case 2, Table III) *B* Type A homogeneous inclusion in the nucleus of an oligoden-

Cont on next page 

a bead-like fashion, along its inner border (Figs. 1A, 1B and 5D). Type A *granular* inclusions vary in size and do not fill the nucleus They are frequently dispersed in irregular clumps. A wide halo often surrounds the inclusion, and beaded marginated basichromatin is usually to be found on the nuclear membrane (Figs 1C and 5E). In the brain the inclusions are eosinophilic, amphophilic or basophilic, while elsewhere, as in the skin and liver, they are usually eosinophilic. Among the structures which are likely to be misinterpreted as inclusions are degenerating plasma cells (Fig 1D).

In a study of the inclusions of herpes simplex by electron microscopy, Morgan and his associates[illegible] noted that the homogeneous inclusions form during an early stage of infection of the cell, with myriad elementary bodies occupying the nucleus, while in granular inclusions elementary bodies are few or absent From these observations they concluded that granular inclusions represent a relatively late stage of inclusion formation The development of inclusions being a dynamic process,[illegible] it is understandable that in any given case there may be forms intermediate between homogeneous and granular inclusions Many of the nerve cells in which inclusions occur are pale and shrunken or otherwise distorted, and in time cytoplasm and nucleus both become more and more ghostlike and disappear.

Type A nuclear inclusions are to be found in the nervous

←- Figure 1—Con't

droglial cell (left), and another in the nucleus of a nerve cell (right) *C* Type A nuclear granular inclusions in nerve cells Peripherally displaced nucleoli are to be seen in *A*, *B* and *C* All X 1460 *D* From a case of B virus encephalitis illustrated in Figure 4, showing a degenerated plasma cell simulating a nuclear inclusion The cytoplasm of the cell is swollen and homogeneous and the nucleus (in the form of a crescent) is displaced to the periphery X 650 *A*-*D*, hematoxylin eosin stain *F* Type B nuclear inclusion in a nerve cell of the anterior horn of the spinal cord in a monkey with poliomyelitis The nucleolus lies beneath the inclusion Hematoxylin-eosin azure stain X810 (Courtesy of Dr A B Sabin, Cincinnati)

system under a number of conditions In herpes simplex encephalitis and in subacute inclusion encephalitis they develop in nerve cells and oligodendroglia, and in the latter cells are found fairly consistently in both grey and white matter Under both conditions, but far more often in the subacute than in the acute form, the cytoplasm of nerve cells contains eosinophilic inclusions of various sizes and shapes which are either single or multiple and only occasionally surrounded by a halo (Fig. 1A). One of us (L. v. B.) has also found characteristic eosinophilic cytoplasmic inclusions in 2 cases of subacute sclerosing leukoencephalitis Differences in the nuclear inclusions in herpes simplex encephalitis and subacute inclusion encephalitis have been described by Foley and Williams.[25] As brought out under *Discussion*, we, too, have observed differences, but they were mostly of a quantitative nature. In chickenpox—herpes zoster, nuclear inclusions sometimes occur in the spinal peripheral nervous system and in the autonomic system (Fig 2) In salivary gland virus disease the cell of the nervous system chiefly affected is the subependymal astrocyte (Fig. 3), and in some cases there may be inclusions in nerve cells of the cerebral cortex as well

Type B inclusions are homogeneous and spherical, single or multiple, and in nerve cells are seldom larger than the nucleolus Nuclear chromatin around the inclusions is destroyed, producing a halo The inclusion does not displace the nucleolus An example of a Type B inclusion from a monkey with poliomyelitis is illustrated in Figure 1E Believing them to be normal chromatin masses, Wolf and Orton[100] questioned their specificity. Hurst[41] then set down characteristics which, he contended, distinguish the two

I. INTERSTITIAL PNEUMONIAS

Giant-Cell Pneumonia with Nuclear Inclusions (Hecht). This disease, a pneumonitis of undetermined etiology—although presumably viral—occurs almost always in children under the age of 2 years[36] Multinucleated giant cells form from the lining cells of alveolar walls and from epithelial cells of alveolar ducts and bronchioles Relatively large granular or homogeneous eosinophilic inclusions indistinguishable from those of herpes simplex

are present in the nuclei not only of giant cells but also of smaller mononuclear cells Eosinophilic inclusions are also to be found in the cytoplasm; they are usually multiple and of various sizes Giant-cell pneumonia with nuclear and cytoplasmic inclusions has been observed in association with measles[7] and whooping cough.[8] It has been suggested that the virus of giant-cell pneumonia may be the same as that of measles[9] We have examined the brain in several cases of giant-cell pneumonia, but have found no inclusions.

Giant-Cell Pneumonia with Cytoplasmic Inclusions (Adams). This form of pneumonia, occurring in children, is much the same as that just described Inclusions are, however, exclusively cytoplasmic, they are present in alveolar lining cells and in giant cells[10] No etiological agent has been found

Virus Pneumonia in Infants (Goodpasture). This form of pneumonia, characterized by the presence of nuclear inclusions in alveolar, bronchial and tracheal epithelial cells, was encountered by Goodpasture and his associates[11] in 5 infants in central Tennessee The inclusions closely simulated those seen in herpes simplex infection An attempt to isolate the virus in one of these cases was unsuccessful Three of the cases occurred in association with measles and a fourth with whooping cough, though in the latter, *Haemophilus pertussis* was not cultured from the lungs, nor could the pulmonary lesions of whooping cough be demonstrated at autopsy In a personal communication to the authors, Dr Goodpasture entertained the possibility that the pneumonia in the infants he and his associates described may have been due to the measles virus

2. CHICKENPOX—HERPES ZOSTER

The viruses of chickenpox and herpes zoster appear to be serologically identical,[r] and resemble each other closely under the electron microscope[n] The inclusions observed in these two diseases are practically identical with those occurring in herpes simplex In studies of infection of human skin grafted on the chorioallantois of chick embryos, Goodpasture and Anderson[*] observed that the nuclear inclusions in herpes simplex infection,

as contrasted with those in *herpes zoster,* become much larger, fill the nucleus, and acquire a basophilic staining *reaction. Size and tinctorial properties are,* however, not always a distinguish*ing feature, for large* basophilic nuclear inclusions as well as shrunken eosinophilic ones have been found *in the skin in herpes* zoster.[illegible]

In chickenpox, inclusions occur chiefly *in* prickle cells *of the epidermis,* though they have been *noted also in the* vascular endothelium and in various cells of the corium.[illegible] The inclusions are nuclear, relatively large, and either eosinophilic or basophilic *They* are *indistinguishable* from the inclusions *of* herpes simplex Smaller ones have been found in the cytoplasm.[illegible]

In occasional instances of chickenpox in infants and young adults (and occasionally in old adults), inclusions develop in macrophages, septal cells and bronchiolar epithelium of the lung in association with pneumonitis, or they are more widespread, occurring variously in the esophagus, pancreas, adrenal, liver, renal pelvis, ureter, and bladder.[illegible] Disseminated lesions limited to viscera and other bodily structures may also occur congenital*ly*[illegible] Inclusions have not been found in the brain in *post* chickenpox encephalitis[illegible]

In herpes zoster in children and adults, the pathological picture of the cutaneous lesions is the same as in chickenpox No inclusions have been found in the central nervous system, although intense inflammatory changes may develop in the posterior root system and posterior grey and white matter of the spinal cord together with sparse lesions in the anterior grey matter medulla oblongata and pons (between the nucleus ambiguus and the spinal nucleus of the Vth nerve and in the nucleus of the VIIth nerve)[illegible] Nuclear inclusions have, however, been observed in the peripheral nervous system

Cheatham[illegible] has found inclusions in the peripheral nervous system of a 50-year-old white woman with Hodgkin's disease who had received a course of X-radiation and some two months later, nitrogen mustard A band like vesiculating rash appeared in the zone of Th 12 on the right side from the umbilicus on to the back, and a prominent vesicular eruption over the entire body A similar band was found on the left Death occurred on the 13th day after the first manifestations of the infection Microscopic examination of

the skin revealed the classical lesions of herpes zoster, with Type A inclusions within epithelial cells in and about the vesicles A few were also seen within neurilemmal cells of small nerve twigs Sections from the esophagus revealed them in cells of the mucous membrane In the stomach, near its junction with the esophagus, neuritis was evident both in the submucosal and myenteric plexuses and at the cardia rather numerous nerve cells of the myenteric plexus contained nuclear inclusions Inclusions of this kind were also found in capsular cells of a dorsal root ganglion and in a sympathetic ganglion at the same level They were also observed in cells of the adrenal medulla, pancreas, and one ovary.

Cheatham and his associates[18] recorded another case in which inclusions were relatively widespread The patient, a 4 year-old white boy, contracted chickenpox while under methotrexate therapy for disseminated neuroblastoma The vesicles continued to appear for 17 days, at which time death occurred Nuclear inclusions were found in the myenteric plexus (Fig 2A), adrenal medulla, capsular cells of the four thoracic dorsal root ganglia which were selected for examination (Fig 2B), and in occasional dorsal root ganglion cells, where they were surrounded by a halo (Fig 2C). Inclusions were also observed in skin and numerous viscera No inclusions were found in the central nervous system

3. SALIVARY GLAND VIRUS INFECTION (CYTOMEGALIC INCLUSION DISEASE)

The causative agent of salivary gland virus infection has now been isolated in human tissue culture in 4 cases of infection in the newborn infant [illegible] This disease occasionally occurs in adults as necrotizing pneumonitis, focal gastrointestinal ulceration, or in a disseminated form [109] In older infants and young children interstitial pneumonia is one of the commonest manifestations of the disease and an association with whooping cough has been frequently reported [illegible] The disease may also occur in young infants in association with interstitial plasma-cell pneumonia, and under such conditions the brain has been found strikingly affected [illegible]

The generalized form of the disease is seen characteristically in newborn and young infants and less frequently in older infants and young children The inclusions, present in the nucleus and frequently in the cytoplasm as well, are found most commonly in epithelial cells of the kidney, liver, pancreas and pituitary gland, and in lining alveolar cells of the lung The affected cells become enormous The nuclear inclusions are correspondingly large,

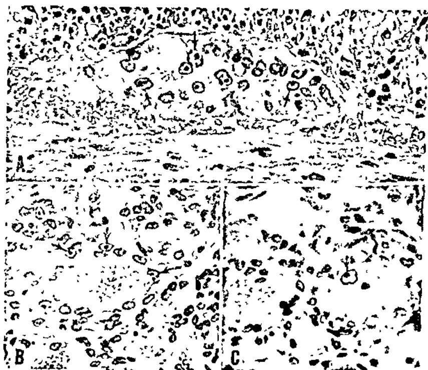


Figure 2 *Chickenpox–herpes zoster, showing nuclear inclusions (A) in nerve cells of the myenteric plexus, (B) in capsular cells around dorsal root ganglion cells, and (C) in one dorsal root ganglion cell Arrows indicate some of the inclusions. In C* ... *ganglion cell shows severe cytoplasmic dege*... eosin stain (Courtesy of Dr ...

either eosinophilic, amphophilic or basophilic, and they often have a granular quality. They are surrounded by a halo, and basichromatin forms bead-like on the nuclear membrane Cytoplasmic inclusions are small and numerous; they are usually basophilic and lack a halo [9,11]

The central nervous system has been affected only in the neonatal period, up to approximately 2 months after birth. The periventricular matrix of the wall of the lateral ventricle (Fig. 3A) and the olfactory tract are the sites of predilection. Calcium and iron salts are deposited in the walls of the lateral ventricles

and are readily visible roentgenographically. The inclusions are present chiefly in astrocytes of the subependymal matrix (Figs 3B and 3C) and less commonly in endothelial and adventitial cells of blood vessels[11] In a case observed by one of us (M. G. S), they were found in numerous nerve cells of the cortex. Polymicrogyria and hypoplasia of the cerebellum have occasionally been observed in this disease[7]

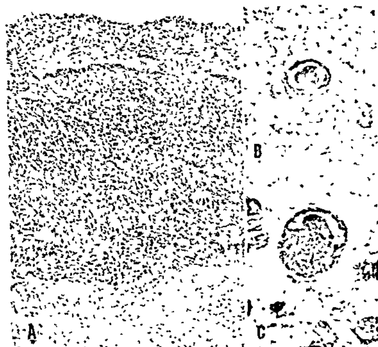


Figure 3 Salivary gland virus encephalitis *A* Great overgrowth of the subependymal matrix of the wall of the lateral ventricle The original surface of the lateral ventricle is indicated by the broken row of ependymal epithelial cells near top of photograph Some of the larger, darker-staining structures in the deeper part of the matrix are inclusion bearing cells X 70 *B* Enlarged subependymal astrocyte, showing nuclear inclusion X 1080 *C* A probable astrocyte showing a large inclusion in the nucleus and myriad tiny inclusions in the cytoplasm X 1080 All stained by hematoxylin eosin (From Haymaker *et al*[7])

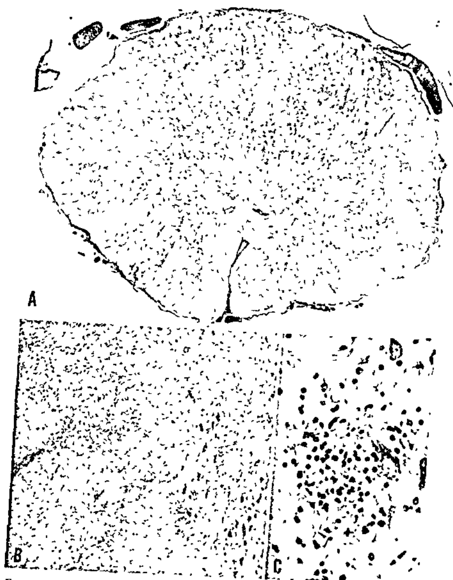


Figure 4 Transverse *myelitis in B* virus infection *A* Lower cervical segment of the spinal cord, showing necrosis of the posterior columns and horns and of the lateral column on one side (to the left) The anterior horns are relatively little affected X 12 Hematoxylin-eosin stain *B* An adjacent

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4. B VIRUS INFECTION

This disease was so named because the surname of the first casualty, a physician working on poliomyelitis, began with the letter B Three fatal cases are known to have occurred

The first infection occurred in 1931 The investigator, aged 29 years, was bitten on the fingers by an apparently normal monkey which harbored the virus in its saliva[80] Death occurred 18 days later Focal necrosis was noted in regional lymph nodes, spleen and adrenals The most striking changes in the nervous system consisted of transverse myelitis at upper thoracic and lower cervical levels, with relatively severe involvement of the posterior columns and horns (in the form of necrosis) (Fig 4A) and pronounced inflammatory

virus after the first letter of the patient's given name.[r]

A second fatal infection with the B virus was reported in 1949[8] While opening a can of penicillin, a physician cut his right index finger He then fed a monkey by stomach tube and contracted the infection from the monkey's

they were easily recognized in the brains of rabbits inoculated with material obtained from an affected right axillary lymph node and central nervous system The virus recovered from this patient was neutralized by a specific B-virus antiserum

A third fatal infection of the same type, following a monkey bite, has recently occurred in a laboratory in Pennsylvania, but has not yet been published[83]

← Figure 4—Con't

spinal segment, illustrating abundant inflammatory cells perivascularly and cells of a similar nature as well as activated glia in the white matter. The

5. ACUTE ENCEPHALITIS IN INCLUSION DISEASE OF THE HERPES SIMPLEX TYPE IN INFANTS UNDER ONE YEAR OF AGE

Disseminated Infection. Herpes simplex viremia accounts for the disseminated lesions of a necrotic and specific nature seen occasionally in young infants. Premature infants are most predisposed. The infection, which can sometimes be traced to exposure of the infant to herpetic vesicles in the mother's vulva, becomes apparent a few days after birth, usually the fifth to seventh day. Disseminated infection may also originate from eruptions in the infant's mouth, conjunctival sac, or skin.[9] Characteristically the liver and the adrenal cortex undergo necrosis, an observation which led Hass,[46] in the first description of the disease in infants, to call the disorder "hepato-adrenal necrosis with intranuclear inclusion bodies." Other structures which may be affected include the spleen, lung, esophagus, stomach, and less commonly, kidney, bone marrow, lymph nodes, and brain. Isolation of the virus from affected organs has been reported in 2 cases,[78, 106] and one of us (M. G. S.) has isolated the virus from the viscera in 2 unpublished cases.

Zuelzer and Stulberg[108] have recounted 8 cases of this form of the disease. Ages at onset were a few days after birth in 5, and between 3 weeks and 32 months in the others. Lesions were widely disseminated in the viscera in all 8 cases. Rather minor changes in the central nervous system were observed in 4 of them.

In Case 1 there were small focal lesions in the medulla oblongata characterized by the presence of invasive mononuclear cells, activated glia and a few leukocytes. Neuronophagic nodules were seen chiefly in the inferior olivary nuclei. In Case 2 a single focus of the same kind was noted in the medulla oblongata, and in Case 3, a similar tiny focus in the pons. Nuclear inclusions were found in nerve cells and glia in Cases 1 and 2 but no reference was made to them in Case 3. In Case 5 the lesion was limited to the cerebral white matter and consisted of necrosis of small blood vessels accompanied by perivascular hemorrhages. No inflammatory cells were observed. Nuclear inclusions were found in a few perivascular glia.

In our series there were 2 cases of disseminated infection of the herpes simplex type (Cases 1 and 2 in Table I). In neither

TABLE 1

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
1 598520 Fetterman	5 days F	5	Full term infant Mother febrile during puerperium & had "influenza" at time infant was born 1st day of illness fever & cyanosis requiring artificial respiration Later lethargy, slt cyanosis, coughed up bloody mucus & died No fever No seizures	—	—	*Gross* Hemorrhagic softening of globus pallidus and cavitation of white matter at angle of lateral ventricle near head of caudate nucleus, both bilaterally *Microscopic* Hepato adrenal necrosis with inclusions
2 796864 WU 17411	7 days (approx) M	8	Premature (7 mo), 1100 gm Day after birth musc twitching & cyanosis when infant was touched or moved 3d day slt jaundice 7th day local or generalized musc twitching on stimulation, episodes of cyanosis 9th day temp 101 5°F, hyperactive reflexes No seizures	Brown red 292 850	3000 (10) 2000 (50)	*Gross* Old subarachnoid blood clot (4 x 1.5 cm) over region of central sulcus focal subarachnoid hemorrhages over hemispheres and elsewhere *Microscopic* Hepato-adrenal necrosis with inclusions In meninges of brain stem, cerebellum and upper spinal cord, severe fibrinonecrotic lymphocytic exudate (Figs 5A & 5B). In cerebral cortex, occasional foci of karyorrhexic nerve cells interspersed with inclusion-bearing nerve cells (Figs 5C 5E) Changes also in basal ganglia and spinal cord (see text)

TABLE I—(*Continued*)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No. AFIP No. Other Nos.	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm.) (% lymphocytes)*	*Pertinent CNS Changes*
3 018707	14 days M	4	Full term infant. At birth slt. lethargy for 1 hr. 1st day of illness cried in "jerky fashion," eyelids & face twitched, many convulsions. 2d day twitching of eyelids & face, tonic musc. contractions in upper limbs & clonic in lower. Later convulsions, temp. 102.4°F.	138	0	*Gross.* Softening of unidentified parts of the cortex. *Microscopic.* Severe damage of striatum, thalamus, pallidum, subthalamus, and adjacent white matter, also of pars basilaris pontis (Fig. 8C) and dentate nucleus of cerebellum (Fig. 8B)
				Xanthochromia CSF pressure "normal"		
4 737141 Kracke[4]	11 mo. M	4	1st day vomiting, convulsions, head held constantly to rt. 2d day attacks (as long as 1 hr.) of musc. contractions of rt. arm & leg & deviation of eyes upward & to rt., temp. 103°F. 3d day generalized convulsions beginning with deviation of eyes to lt., hemiparesis on rt. 4th day death following repeated convulsions.	—	60	Gross: Softening of hippocampal region, insular region, fronto parietal cortex and gyrus cinguli, side unknown. *Microscopic.* In temporal cortex, abundant foci (sometimes laminar) of rarefaction necrosis (Fig. 7), and in white matter, multiple regions of foci of necrosis, many coalescent (Fig. 7); and in superolateral part of the parietal and occipital lobes, demyelination and focal necrosis in the superficial and deep white matter. Similar changes in gyrus cinguli
				No bulging of fontanelle		

TABLE I—(Continued)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No AFIP No Other Nos	Sex Age at Onset	Duration of Illness (days)	Clinical Features	Spinal Fluid Protein (mg %)	Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
				Other Data		
5 796859 WU 6154	6 mo F	6	1st 4 days listless, irritable feverish generalized convulsions lasting 5 hrs 5th day unconscious slt spasticity	—	150	*Gross* Meninges and brain edematous No brain changes *Microscopic* Minimal lesions in cerebral cortex, thalamus, pons and dentate nucleus
6 748020	11½ mo M	6	1st 3 days vomiting, lethargy, fever 4th day temp 102°F, convulsions involving rt side purposeless jerking mvts of rt arm & leg spreading to lt arm & leg coma 5th day head & eyes constantly rotated to lt, all limbs spastic, espec rt 6th day convulsion neck stiff, severe carpopedal spasms	36 80 CSF pressures 250 & 260 mm H_2O Papilledema lt, rt disc not seen Brain bulged on trephine for suspected subdural hematoma	5 (100) 9 (78)	*Gross* No changes except for brain swelling *Microscopic* Characteristic necrosis in temporal cortex and insula and adjacent white matter Rather little change elsewhere

TABLE I—(Continued)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Other Data*	*Pertinent CNS Changes*
3 618707	14 days M	1	Full-term infant At birth, slt lethargy for 1 hr. 1st day of illness cried in "jerky fashion," eyelids & face twitched many convulsions 2d day twitching of eyelids & face, tonic musc contractions in upper limbs & clonic in lower Later convulsions, temp 102 4°F	138	0	Xanthochromia CSF pressure normal	*Gross.* Softening of unidentified parts of the cortex *Microscopic* Severe damage of striatum, thalamus, pallidum, subthalamus, and adjacent white matter, also of pars basilaris pontis (Fig 8C) and dentate nucleus of cerebellum (Fig. 8B)
4 757141 Krucke[4]	11 mo M	1	1st day vomiting, convulsions head held constantly to rt 2d day, attacks (as long as 1 hr) of musc contractions of rt arm & leg & deviation of eyes upward & to rt, temp 103°F 3d day generalized convulsions beginning with deviation of eyes to lt hemiparesis on rt. 4th day death following repeated convulsions	—	60	No bulging of fontanelle	*Gross* Softening of hippocampal region, insular region, fronto parietal cortex and gyrus cinguli, side unknown *Microscopic* In temporal cortex, abundant foci (sometimes laminar) of rarefaction necrosis (Fig 7), and in white matter, multiple regions of foci of necrosis, many coalescent (Fig 7), and in superolateral part of the parietal and occipital lobes, demyelination and focal necrosis in the superficial and deep white matter. Similar changes in gyrus cinguli

TABLE I—(Continued)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No AFIP No Other Nos	*Sex Age at Onset*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
5 796959 WU 6154	6 mo F	6	1st 4 days listless irritable feverish generalized convulsions lasting 5 hrs 5th day unconscious slt spasticity	—	130	*Gross* Meninges and brain edematous No brain changes *Microscopic* Minimal lesions in cerebral cortex, thalamus, pons, and dentate nucleus
6 748020	11½ mo M	6	1st 3 days vomiting lethargy, fever 4th day temp 102°F convulsions involving rt side, purposeless jerking mvts of rt arm & leg spreading to lt arm & leg coma 5th day head & eyes constantly rotated to lt all limbs spastic, espec rt 6th day convulsion neck stiff, severe carpopedal spasms	36 80	5 (100) 9 (78)	*Gross* No changes except for brain swelling *Microscopic* Characteristic necrosis in temporal cortex and insula and adjacent white matter Rather little change elsewhere
				CSF pressures 250 & 260 mm H_2O Papilledema, lt., rt disc not seen Brain bulged on trephine for suspected subdural hematoma		

TABLE I—(Continued)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN 9 INFANTS UNDER ONE YEAR OF AGE

Case No. AFIP No. Other Nos.	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
7 796863 W U 17449	13 days M	8	Full term infant. 1st day of illness irritable; 2d day seizures (5-6 times each 10-15 min) consisting of twitching of lt eyelid, lt side of face & lt arm & hand, subsequently incl lower limbs with tonic & clonic phases. Later (in hosp), temp 99.2°F, repeated convulsions, coma	173	2500 (5)	*Gross* Hemorrhagic softening, temporal lobes (especially fusiform gyrus) and orbital cortex, bilaterally, less severe in other lobes, with sparing of rolandic region and caudal part of occipital lobe. *Microscopic* Cortical necrosis in temporal, superior frontal and occipital regions (Fig 6A) with proliferating subpial astrocytes. Hippocampal formation, severely affected bilaterally, with numerous inclusions (Fig 8D). Thalamus, occasional focal necrosis and large collections of perivascular and invasive cells; striatum, less affected; subthalamic nucleus, striking porosity and occasional invasive cells; inferior olivary nucleus, nerve-cell loss with several large foci of perivascular invasive cells

	Other Data	
7	Bulging ant fontanelle, diastasis of cranial sutures No herniation through incisura cerebelli post mortem	

TABLE I—(Continued)

ACUTE ENCEPHALITIS OF THE HERPES SIMPLEX TYPE IN INFANTS UNDER ONE YEAR OF AGE

Case No. AFIP No. Other Nos.	Age at Onset Sex	Duration of Illness (Days)	Clinical Features	Spinal Fluid: Protein (mg %)	Spinal Fluid: Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
8 797041 W U 8095 Smith Lennette & Reames*	1 mo M	8	Premature infant (8 mo) 1st day fretful & irritable Later twitching of lt arm & leg which became continuous, one generalized convulsion slt fever, slt clouding of cornea	Bloody Fontanelle tense No papilledema		*Gross* Much of brain softened, with prominent involvement of "olfactory areas" Most of cerebrum discarded because soft and mushy *Microscopic* Striking changes in lower oblongata (Fig 8A) (see text)
9 796861 W U 16486	7 days F	19	Full term infant 1st day of illness irritable 3d day convulsion 6th day opisthotonos, spasticity Later frequent convulsions, semicoma, spastic, pupils nonreactive conjunctivitis Temp remained subnormal	— 410	155 (93) 115 (93)	*Gross* Entire brain soft and mushy, petechiae in meninges Opaque white plaques" in cerebellar meninges *Microscopic* Severe changes in cerebral cortex, basal ganglia, anterior perforated substance, midbrain (Fig 6B), pons and medulla oblongata

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(*Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection*)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (Days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
3 109395 Whitman *et al* Haymaker	26 yr M	7	1st day sudden severe headache 2d day headaches, vomiting 3d day fever, irrational Later confused, disoriented, temp 101 8°F, drowsy, neck stiff, rt leg spastic, rt arm flaccid, rt pupil dilated and fixed, coma No seizures	93 106	700 (97) 1010 (97) Many RBC	*Gross* Brain "soft and mushy," 2 5 cm area of softening in inferior aspect of rt. occipital lobe Much of cerebrum discarded because softened *Microscopic:* Temporal cortex, characteristic breakdown (Fig. 10A), enlarged astrocytic nuclei in all laminae, subpial astrocytic hyperplasia, and neuronophagia (Fig 13A). Rt insular and occipital cortex similarly affected Mainly nerve-cell necrosis in basal ganglia
4 615507	31 yr M	8	For previous 6 mo undue thirst and personality change Felt something wrong in head "for long time". Often talked of dying 1st and 2d days of present illness: head ache, chills, confusion, temp 101°F. 7th day, no jacksonian seizure, agitated, no localized weakness, coma	64 No papilledema Ventriculogram shift of lt hemisphere to rt.	260 (100)	Biopsy of lt temporal and frontal lobes diagnosed "acute cerebritis" *Gross:* Lt temporal lobe necrotic and diffusely hemorrhagic. *Microscopic* Only slight changes in basal ganglia and brain stem. Around caudate nucleus and thalamus, white matter demyelinated and contains many perivascular inflammatory cell cuffs and invasive ameboid cells

TABLE II—(Continued)
ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (Days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
5 *796865* WU 10298	14 yr M	8	1st day severe headache 2d day fainted, vomited 3 times, temp 101°F 4th day dazed, convulsion Later stiff neck, stupor, coma, temp 105°F. No skin eruption, but brother (his bedfellow) had had recurrent herpes labialis, which was present at the time this illness began	— — — No papilledema	200 290 420 (mostly lymphocytes)	*Gross* Softening of mesial part of temporal lobes, mostly on lt, spongy area (7 5 cm) in floor of posterior horn of lt lateral ventricle *Microscopic.* In temporal cortex, characteristic breakdown in some regions, multiple glomesenchymal cell nodules in others (Fig 131) In anterior nucleus of thalamus, large perivascular cell cuffs; in pulvinar, sparse cell cuffs Other basal ganglia relatively little affected
6 *85691* Zarafonetis *et al*[104] Haymaker[46]	25 yr M	8	1st day headache, fever, weakness, confusion 3d day disoriented, delirious, temp 103 8°F, neck stiff, anisocoria, hiccup, rt hemiparesis, ptosis of rt eyelid Later delirium, opisthotonos, temp 105° F No seizures	44 50 CSF pressure 110 mm H_2O No papilledema (2 exam's) Gold curve flat	175 (98) 323 (97)	*Gross* In lt. temporal lobe, area of softening (4 cm in diameter) studded with hemorrhages *Microscopic* Characteristic necrosis of temporal cortex and damage to subcortical white matter In pulvinar, many necrotic nerve cells

Other Data

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)

(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
16 789857 IB 107/57 Kleyntjens	17 yr M	12	Chronic alcoholic 1st day agitation followed by unconsciousness and lt hemiplegia 2d day temp 99 6°F, semi coma 4th day stuporous, temp 100°F Later remained comatose, eyes tended to deviate to rt, many convulsions	20 10 No papilledema	72 (80) 17 (25)	*Gross* No data *Microscopic* In temporal cortex (only 2 blocks available), most areas show severe damage of lower or of all laminae with large collections of inflammatory cells around many vessels; virtually total breakdown of cortex in walls of some sulci In white mat- e section massive break- l multiple cavity forma- here severe and extensive ith disappearance of glia, ory cells around numer- , macrophages both peri- ind diffuse, many small tes
17 757143 175984 P 551 Adams	77 yr M	12	1st day rose in middle of night, dressed, shaved, told his wife he was going to a funeral 3d day headache, vomiting Later, found in street unconscious, tremulous, disoriented but jovial, slt stiffness of neck, trephine No seizures Cutaneous lesions of lt, frontal region and nose (nature?)	Fluid Clear WBC 200 RBC 120 Xanthochromia RBC 1230 RBC 3600 CSF Pressures 200, 125 and 120 mm H_2O		tening, rt temporal lobe ; hippocampal formaus cinguli, thalamus, and ivision of pallidum *Mic-* Necrosis, with character- mmatory-cell reaction, in Indicated in gross exami- White matter of frontal gyrus cinguli, thalamus nigra, red nucleus and oblongata, perivascular [illegible] invasive cells

TABLE II—(Continued)
ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness (days)	Clinical Features	Spinal Fluid Protein (mg %) / Other Data	Spinal Fluid Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
18 *660941*	30 yr M	12	1st day became wet in rain, headache, fever, chill 2d day vomiting 5th day delirious, disoriented, temp 101 6° F, tracheotomy because of respiratory difficulty Later hiccup, pos grasp reflex, rt hemiparesis, twitching of ms of rt hand, foot and face, tremor of rt hand, horizontal nystagmus, stiff neck, ecchymoses in sclerae, stupor No seizures	72 78 Xanthochromia No papilledema CSF pressure 500 mm H_2O Gold curve flat Ventriculogram shift of rt. hemisphere to lt Severe herniation of both hippocampal gyri post mortem	88 (95) 161	Gross Hemorrhagic softening of rt temporal lobe (including uncus) and insular region, less marked on lt. *Microscopic.* In frontal cortex, scattered focal rarefaction and laminar necrosis with occasional groups of rod cells and enlarged astrocytic nuclei In occipital cortex, inflammatory-cell cuffs. In sublenticular region, anterior perforated substance and amygdala, focal perivascular and diffuse tissue breakdown. Rt lenticular nucleus, moderately severe inflammatory changes, thalamus, slight In hypothalamus (mamillary level), many scattered cell cuffs and invasive cells, rare in mamillary body
19 325161	45 yr F	13	1st day felt cold, temp 99 2° F 3d day dysarthria, stuttering, unsteady gait, titching of eyelids, could write words but not pronounce them, rt hemiparesis, severe edema of uppr lip and lt upper eyelid, many rt facial clonic seizures spreading to rt neck and chest (and sometimes to rt arm) with unconsciousness, palatal palsy, temp 101 6° F	40 — No papilledema CSF pressure 390 mm H_2O	33 (66) 110	*Gross: Hemorrhagic* softening in region of lt basal ganglia, less on rt, small hemorrhagic area "lateral to striatum". *Microscopic* In temporal and cingulate cortex, characteristic necrosis In cerebral white matter in region of damaged cortex, subcortical perivascular cell cuffs, and, in places, perivascular tissue breakdown In putamen, little change

TABLE II—(Continued)

Acute Onset and Rapid or Prolonged Course. Acute Inclusion Encephalitis (37 Cases)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
16 788857 IB 107/57 Kleyntjens	47 yr M	12	Chronic alcoholic 1st day, agitation followed by unconsciousness and lt hemiplegia 2d day temp 99 6°F, semi coma 4th day stuporous, temp 100°F Later remained comatose, eyes tended to deviate to rt, many convulsions	20 10 No papilledema	72 (80) 17 (25)	*Gross.* No data. *Microscopic* In temporal cortex (only 2 blocks available), most areas show severe damage of lower or of all laminae with large collections of inflammatory cells around many vessels, virtually total breakdown of cortex in walls of some sulci In white matter, in one section massive breakdown and multiple cavity formation; elsewhere severe and extensive necrosis with disappearance of glia, inflammatory cells around numerous vessels, macrophages both perivascular and diffuse, many small hemorrhages
17 757145 175984 P-551 Adams	77 yr M	12	1st day rose in middle of night, dressed, shaved, told his wife he was going to a funeral 3d day headache, vomiting Later, found in street unconscious, tremulous, disoriented but jovial, slt stiffness of neck, trephine No seizures. Cutaneous lesions of lt frontal region and nose (nature?)	Fluid Clear WBC 200 RBC 120 Xanthochromia RBC 1230 RBC 3600 CSF Pressures 200, 125 and 120 mm H_2O		*Gross* Softening, rt temporal lobe (including hippocampal formation), gyrus cinguli, thalamus, and external division of pallidum *Microscopic* Necrosis, with characteristic inflammatory-cell reaction, in structures indicated in gross examination. White matter of frontal lobe and gyrus cinguli, thalamus substantia nigra, red nucleus and medulla oblongata perivascular and occasional invasive cells

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)

(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Other Data*	*Pertinent CNS Changes*
18 *660841*	30 yr M	12	1st day became wet in rain, headache, fever, chill 2d day vomiting 5th day delirious, disoriented, temp 101 6°F, tracheotomy because of respiratory difficulty. Later hiccup, pos grasp reflex, rt hemiparesis, twitching of m's of rt hand, foot and face, tremor of rt hand, horizontal nystagmus, stiff neck, ecchymoses in sclerae, stupor No seizures	72 78	89 (95) 161	Xanthochromia No papilledema CSF pressure 300 mm H_2O Gold curve flat Ventriculogram shift of rt hemisphere to lt Severe herniation of both hippocampal gyri post mortem	[illegible] oftening of [illegible] ding uncus) [illegible] marked on [illegible] ntal cortex, [illegible] on and lami- [illegible] ional groups [illegible] ed astrocytic [illegible] tex, inflam- [illegible] ublenticular [illegible] orated sub- [illegible] focal peri- [illegible] issue break- [illegible] cleus, mod- [illegible] ory changes, [illegible] pothalamus [illegible] ny scattered [illegible] cells, rare
19 325161	43 yr F	13	1st day felt cold, temp 99.2° F 3d day dysarthria, stuttering, unsteady gait, twitching of eyelids, could write words but not pronounce them, rt hemiparesis, severe edema of uppr lip and lt upper eyelid, many rt facial clonic seizures spreading to rt neck and chest (and sometimes to rt arm) with unconsciousness, palatal palsy, temp. 104 6° F	40 —	33 (66) 110	No papilledema CSF pressure 390 mm H_2O	[illegible] oftening in [illegible] glia, less on [illegible] rea "lateral [illegible] ne In tem- [illegible] ortex, char- [illegible] rebral white [illegible] imaged cor- [illegible] scular cell- [illegible] perivascular [illegible] tamen, little

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(*Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection*)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)* / *Other Data*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
20 312131	72 yr M	13	During 1 mo after head injury, confusion, memory lapses nausea and dysuria Then 20 'moles' were fulgurated from face Became ill next day aching m's, chills and fever 4th day nausea, temp 102° F 7th day convulsion, coma stiff neck, eyes deviated to rt	64 116 33 CSF Pressure 210 mm H_2O Gold 0001110000	73 (100) — 2	*Gross* Softening (with petechiae), lt temporal pole, hippocampal formation, insula, anterior perforated substance, and orbital gyri. Rt side practically spared *Microscopic* In parieto occipital cortex, abundant perivascular inflammatory cells and slight tissue breakdown with ameboid cell invasion of nervous tissue, occasional cell-nodules, foci of rod cells, and scattered enlarged astrocytic nuclei In cerebral white matter, little change except for hemorrhages near insula
21 289115	55 yr M	14	1st 3 days slt nausea 4th day nausea, vomiting, climbed fence and attempted to push neighbor's house down with hands, talked foolishly, 'some' convulsions, stupor, neck stiff, eyes turned to lt Later clonic contraction of lt arm with head and eyes turned to lt, nystagmus temp 103° F	28 187 — CSF pressure 170 mm H_2O Gold curve flat	36 (100) 8 (100) 18 (100)	*Gross* Anterior inferior aspects of both temporal lobes softened and hemorrhagic, mostly on rt, to a depth of 2.5 cm *Microscopic* In temporal cortex, varied picture ranging from acute lesions unattended by reaction to chronic sclerosing lesions, with proliferative astrocytic reaction extending deep into white matter (for details see pages 153 and 154, in small print) Sublenticular region, several large cuffs of lymphocytes and widespread astrocytosis No significant change in thalamus caudate nucleus or globus pallidus

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
22 757144 IB 155 55	47 yr F	14	1st day nausea, vomiting sore throat, slt icterus 2d day unconscious Later coma lt hemiparesis, generalized musc rigidity, trismus, palatal palsy, strabismus, twitching of m s in rt face and foot frequent rt jacksonian seizures (often rhythmic) many convulsions, temp 105°F	80 100 90 90 70 No papilledema (2 exams') retinal hem s	170 (100) 600 (100) 800 (90) 1100 (81) 900 (77)	*Gross* Softening, rt temporal lobe *Microscopic* In cerebral white matter (beneath damaged cortex) numerous perivascular foci of "ischemic change," especially at depths of sulci, scattered perivascular foci of tissue damage, slight myelin pallor in temporal and orbital regions with enlargement of astrocytic nuclei
23 726491 IB 119/55 Cordier & Henneaux[19]	57 yr F	14	For 6 mo, involutional psychosis Onset of present illness 48 hr after 4th electro shock treatment fever, somnolence, stupor, stiff neck, totally inert, intestinal atonia diarrhea 11th day vascular collapse (shock), temp 101°F No seizures	22	4	*Gross* Softening, rt temporal lobe, insular region, gyrus cinguli and caudate nucleus *Microscopic* In temporal cortex, necrosis and widespread tissue breakdown, pronounced astrocyte enlargement, mostly subpially In cerebral white matter, myelin pallor (with minor perivascular cell collections) in the ventral part of the centrum semiovale, temporal pole, gyrus cinguli, and region of basal ganglia Putamen shows large perivascular cell-cuffs laterally, contiguous with necrotic claustrum and white matter

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE: ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
24 796866 WU-13943	21 yr F	14	1st day headache. 2d day confused, had convulsion, vomited 3d day stuporous 8th day: purposeless mvts, hippus, pos grasp reflex, constant deviation of eyes to lt, flaccid paralysis, coma Tracheotomy. 11th day temp 104°F, small grey ulcer on lt buccal mucous membrane, which "might be a herpetic lesion"	131 135 No papilledema CSF pressures: 220 and 250 mm. H_2O Gold curve flat	250 (98) 571 (98)	*Gross* Softening, both temporal lobes (including uncus and hippocampal gyrus) and of anterior perforated substance, mostly on rt, also of insula and gyrus rectus, mostly on lt. *Microscopic*: In temporal lobes, characteristic cortical necrosis, scattered rod cells, many enlarged astrocytic nuclei, focal tissue breakdown with macrophage response; perivascular inflammatory-cell reaction in subcortical and deep white matter, with pleomorphic cells invading deep white matter, many fibrillary astrocytes In orbital gyri, severe damage of both grey and white matter. In lt. frontal and lt. parietal cortex, scattered cell nodules and perivascular lymphocytes Lt. occipital lobe spared In anterior commissure, substantia innominata and anterior perforated substance, numerous large perivascular cell-cuffs, pleomorphic invasive cells, fibrillary astrocytic hyperplasia, and rod cells Hypothalamus (post and lat areas and mamillary bodies), somewhat less affected Thalamus and subthalamus, still less Globus pallidus, large cell cuffs laterally

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No. AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features* [illegible]	*Spinal Fluid Protein (mg. %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Other Data*	*Pertinent CNS Changes*
25 614688	22 yr M	14	1st 4 days slt then severe headache, persistent nausea, vomiting, no fever *5th day* temp 101 4°F, "peculiar smell of room," slt confusion, hiccup, severe frontal headache Later: disorientation, hippus, stiff neck, frequent convulsions, repeated jacksonian seizures in lt face and arm (every 20 30 min) often becoming generalized	34 140 172	238 (95) 616 (100) 133 (100)	No papilledema	*Gross* Rt temporal lobe extremely soft and studded with hemorrhages Smaller areas (1 2 cm) of softening in rt. parietal, occipital and precentral cortex *Microscopic.* In temporal cortex, acute thrombophlebitis in meninges with necrosis of adjacent cortex, many inclusions in this region, multiple small areas of rarefaction necrosis, scattered rod cells, slight astrocyte enlargement In subcortical region of white matter, many perivascular cell cuffs and tissue breakdown Little change elsewhere
26 336038	26 yr M	15	Previous 3 wk tired, had "far away look in his eyes and looked weak". 1st day of present illness malaise, sleepy 3d day, headache, convulsion Later many convulsions, coma, aniscoria, periodic deviation of rt. or both eyes to rt, twitching of facial m's, laryngeal spasms, musc rigidity, ventriculography (results not known), temp to 106°F	75 90	220 (95) 158 (16)	Xanthochromia CSF pressure 120 mm H O *Gold.* 5344332110 5555322210	*Gross* Rt. hemisphere swollen, hemorrhagic softening in anterior 2/3 of rt. temporal lobe and in hippocampal formation. Same but less on lt *Microscopic* In temporal and cingulate gyri, characteristic necrosis, hyperplastic astrocytes in and near necrotic foci, occasional foci with only nerve cell karyorrhexis, occasional rod cells In cerebral white matter, multiple patchy necrotic areas containing hyperplastic astrocytes and scattered perivascular cell cuffs. In hypothalamus, many perivascular cell cuffs dorsally at tuberal level, small focus of tissue necrosis in mamillary body, Basal ganglia, little change

TABLE II–(Continued)

Acute Onset and Rapid or Prolonged Course. Acute Inclusion Encephalitis (37 Cases)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness (days)	Clinical Features	Spinal Fluid Protein (mg %)	Spinal Fluid Cells (cmm) (% lymphocytes) Other Data	Pertinent CNS Changes
27 708825 IB 83 53 v Bogaert Gambier & Henneaux	54 yr M	15	Grippe with epigastric pain, fever, then somnolence confusion, muscular rigidity, coma No seizures	29	172 (100)	*Gross* Softening of one temporal lobe (including hippocampal gyrus), insular region, gyrus cinguli, parts of thalamus, caudate nucleus, lat geniculate bodies, and optic nerves *Microscopic.* In white matter, in region of temporal pole and in orbital and subcallosal gyri, intense pallor with scattered cell-cuffs, occasional perivascular tissue damage, and enlarged astrocytic nuclei
28 706858 WU 11969	25 yr F	16	1st day severe headache, aching knees, felt "quivery" 7th day weakness, espec rt hand 8th day vomiting Later lethargy, stiff neck, lt arm rigidly flexed, coma, temp 102 6° F. No seizures	215 320 — No papilledema CSF pressures "normal", 215, 165, 155 mm H₂O	268 (81) 914 (75) 308	*Gross* Rt temporal lobe softened and studded with petechiae. Lt, less softened *Microscopic* In lt parolfactory and orbital gyri, necrotic foci in upper laminae, many cell nodules, and much perivascular inflammatory reaction, underlying white matter little affected In rt, superior frontal gyrus, many pleomorphic cell nodules, numerous enlarged astrocytic nuclei, occasional perivascular cell cuffs and a few foci of rarefaction necrosis, in underlying white matter, a few large cell cuffs and rare cell nodules Lt frontal parietal cortex, spared Thalamus, moderately affected

TABLE II—(Continued)
ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness (days)	Clinical Features	Spinal Fluid Protein (mg %)	Spinal Fluid Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
29 671011	30 yr M	17	1st day eyes felt like they were closing Next 3 days feverish headache and retrobulbar pain confusion, delirium temp 104°F neck rigid, lt pupil fixed rt reacted normally Later coma divergent strabismus No seizures	112 145 Other Data: CSF pressure 210 mm H_2O	197 (81) 113 (87)	*Gross* Hemorrhagic softening, rt temporal lobe, both insulae and rt hippocampal formation and amygdala Relatively large area of hemorrhagic softening in lt putamen and adjacent white matter *Microscopic* In temporal cortex lymphocyte and plasma cell infiltrates widespread astrocytic and patchy rod cell proliferation and nerve cell loss (Fig 15A), also neuronophagia (Fig 15C), gliomesenchymal nodules (Fig 15B), and, in subcortical white matter, perivascular gliomesenchymal reaction (Fig 17A) Similar changes in other areas of cortex, including occipital Putamen, caudate nucleus globus pallidus and lat geniculate body, affected decreasingly in that order Little involvement of other basal structures

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(*Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection*)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes) Other Data*	*Pertinent CNS Changes*
30 299725	27 yr M	18	1st day severe throbbing frontal headache 2d and 3d days fever, slt stiff neck 4th day convulsion, semistupor In hosp at end of 2d week, temp 102°F, stiff neck, stupor	109 118 —	355 (100) 28,575 (RBC 80% WBC 20%) 183,000 (RBC 88%, WBC 12%) Xanthochromia No papilledema (2 exam's) Gold. 5554322100	*Gross* Copious hemorrhages over anterior 1/3 of rt. temporal lobe with 6 cm. area of softening, same on lt, 3 cm. in diameter. *Microscopic.* In temporal cortex, multiple large hemorrhages associated with thrombosed vessels, characteristic focal tissue breakdown, widespread nerve-cell necrosis, enlargement of astrocytic nuclei. Micronodule ("*Gliaknotchen*") type of encephalitis in some fields In cerebral white matter, in occasional areas a few large cell-cuffs and scattered invasive cells beneath relatively intact cortex. Wall of lateral ventricle, many huge perivascular cuffs of lymphocytes Pulvinar, severely affected Rather little change in other cerebral structures

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)

(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
31 708823 IB 108/54 Recordier et al[18]	50 yr F	20	First few days fever. Later, agitated, comatose, stiff neck, automatic mvts, bilat myoclonic contractions of eyelids, lips and upper limbs (espec on lt), head deviated to rt, pos grasp reflex on lt No convulsions	38 80 170 104 150 110 88 112 112 60 62	26 (100) 175 (91) 450 (72) 396 (83) 286 (98) 93 (93) 145 (46) 214 (99) 230 (89) 298 310 (30)	*Gross*, Softening, both temporal lobes (especially hippocampal gyrus) (mostly on rt), rt gyrus cinguli, nucleus accumbens septi, orbital gyri, and both amygdalae *Microscopic* In cerebral cortex, characteristic necrosis Myelin pallor, with sparse cell infiltrates, in temporal pole and orbital and cingulate gyri Cell infiltrates also in white matter of frontal and parietal lobes and in septum pellucidum, thalamus, putamen, and anterior part of corpus callosum
32 230630	28 yr M	20	1st day severe nuchal pain, nausea 9th day *déjà vu* attacks loss of memory for recent events, forgetful, tremulous 14th day temp 102°F, sudden stiffness of body with unconsciousness for several minutes, then spoke jargon, insensitive to pinprick Later stiff neck, many convulsions	Xanthochromia CSF pressure 280 mm H_2O		*Gross* Brain "gelatinous" Softening (with petechiae) of lt temporal lobe (especially anterolateral and basal parts) and adjacent region of occipital and parietal lobes *Microscopic* In temporal cortex, characteristic focal tissue breakdown and, in deeper laminae, extensive rarefaction necrosis, in some areas, numerous hypertrophied astrocytes In temporal white matter, severe patchy tissue damage (with numerous perivascular foci of invasive cells) extending deep into white matter In putamen, rather numerous large cell cuffs and occasional foci of invasive cells

TABLE II—Continued

Acute Onset and Rapid or Prolonged Course. Acute Inclusion Encephalitis (37 Cases)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No. AFIP No. Other Nos.	Age at Onset Sex	Duration of Illness (days)	Clinical Features	Spinal Fluid Protein (mg %)	Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
				Other Data		
33 *480864*	56 yr M	21	1st wk confused could write but not utter sounds 8th day (in hosp) frequent attacks (lasting 1/2 hr) of epigastric pain, lacrimation (lt eye) salivation and sweating of hands, bulbar palsy (lt 7th 9th and 10th n's and rt 12th n) and intercostal paralysis, anisocoria lt hemiparesis Later twitching of leg m's, stiff neck frequent jacksonian seizures, temp around 103°F	101 — Gold 1111000000	53 (6) 12 (80)	*Gross* 'Parietal' cortex contained 3 hemorrhagic areas 2 cm in diameter on surface and 3 cm deep *Microscopic* Meninges show abundant coagulated edema fluid and characteristic cell collections. In temporal cortex, characteristic necrosis, marked astrocytosis in uppermost lamina and enlarged astrocytes through all laminae and intensely hemorrhagic areas associated with thrombi In temporal white matter, pronounced subcortical rarefaction necrosis, with scattered perivascular foci of invasive cells Little involvement of basal nuclei Insufficient sections to determine basis of palsy

TABLE II—(Continued)

Acute Onset and Rapid or Prolonged Course: Acute Inclusion Encephalitis (37 Cases)
(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness (days)	Clinical Features	Spinal Fluid: Protein (mg %)	Spinal Fluid: Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
				Other Data		
34 697229 IB 117 58 Bogaert Radermecker & Devos[38]	22 yr M	25	1st day sore throat after exposure to cold confused 2d day stiff neck temp 101°F, somnolent Later disoriented agitated signs of lt cerebello tegmental damage, deviation of head and eyes to lt flaccid quadriplegia, convulsive mvts starting in lt hand and advancing to involve entire body	22 CSF pressure 140 mm H O Gold 0002210000	264 (95)	*Gross* Softening of much of temporal lobe (including hippocampal gyrus uncus and amygdala), insula, anterior perforated substance, orbital cortex and septal region, gyrus cinguli, thalamus and lenticular nucleus, all most severe on lt *Microscopic* Temporal cortex, characteristic necrosis In subcortical and occasionally in deep white matter large perivascular collections of ameboid cells with numerous cells of a similar nature in surrounding tissue (Fig 17C), and myelin pallor in temporal pole and in region of cingulate and orbital gyri and internal capsule
35 777215 IB 102 56	36 yr M	15	1st 3 wk vomiting vague visual disturbances Later strabismus temp 100°F confusion violent headache neck rigid incessant vomiting, agitation convulsions auditory and visual hallucinations lt hemiparesis lt hyperpathia	10 60 90 110	72 (60) 84 310 330 (95)	*Gross* Softening, temporal lobe (including hippocampal formation), bilaterally *Microscopic* In temporal lobe necrosis with upper laminae replaced by masses of ameboid cells and microphages also perivascular macrophages, rather few hypertrophic astrocytes enormous collections of perivascular cells in cortex and adjoining white matter, with scattered foci of necrosis in subcortical white matter scattered perivascular cells in white matter of temporal lobe hippocampal formation and cingulate gyrus

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)

(Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
36 711883 IB 62 54 Berger	65 yr M	3½ mo	Previous few mo forgetful, lost 14 lb in weight 1st day of present illness frontal headache 4th day confused, disoriented Later 3 episodes of coughing, cyanosis and loss of consciousness, temp 101°F, rt hemiparesis, dysphagia 2d mo could sit up in bed Later salivation, incoherent When about to be discharged from hosp had convulsive seizure and died	52 Xanthochromia No papilledema	—	*Gross* Severe softening of lt temporal lobe (including hippocampal formation), gyrus cinguli and orbital cortex. *Microscopic:* Temporal lobe, cavitation of most of cortex with breakdown of white matter (Fig 16) Inferior parietal cortex, sclerotic, with tissue breakdown at base of one deep sulcus and in insula Frontal cortex, devastation at base of occasional sulcus; elsewhere, many cell nodules and much sclerosis Occipital cortex, sclerosis and cell-nodules in medial and inferior cortex and occasionally in lateral cortex, calcarine region spared Unidentified cortex, curious hyperplastic symplasmatic-like adventitial cells with pleomorphic nuclei and abundant cytoplasm Gyrus cinguli, destroyed Thalamus, medial third shows many pleomorphic reactive cells in interstitium Caudate nucleus, sclerosed Other basal structures show little change

TABLE II—(Continued)

ACUTE ONSET AND RAPID OR PROLONGED COURSE ACUTE INCLUSION ENCEPHALITIS (37 CASES)
(*Italicized AFIP Numbers Indicate Proven Herpes Simplex Infection*)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness (days)*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Spinal Fluid Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
37 597473 Dodge & Cure	20 yr M	4 yr (still living)	Acute onset headache, vomiting, confusion disorientation temp 103°F Treated as case of heat exhaustion Later neck rigid rt hemiparesis choreoathetosis of lt upper limb frequent hiccup coma Not seizures Craniotomy	111 CSF pressure 355 mm H_2O No papilledema Ventriculogram shift of rt hemisphere to lt Severe herniation of brain on decompression	188 (67)	Biopsy, softened rt temporal lobe *Microscopic* Rt temporal, parietal and occipital lobes (on 3d day of illness), perivascular necrosis in cortex with abundant ameboid cells Numerous nuclear inclusions

Other Data (listed under Spinal Fluid Protein)

the neck and muscular weakness and flaccidity or spasticity were common. Hemiparesis or hemiplegia was noted in 12 cases, and even more frequently *a positive Babinski response unilaterally or* bilaterally. Acute esophageal ulceration with hemorrhage was observed post mortem in one case (Case 24).

Seizures of one kind or another occurred in 24 cases: convulsive (19 cases), jacksonian (9), and psychomotor (1; Case 33). Hallucinations were complained of in 4 (Cases 10, 12, 25 and 35), and *déjà vu* episodes in 1 (Case 32), the latter consisting of rapid recollection of all illnesses during the previous 10 years One patient had attacks of sudden stiffness of the body and unconsciousness for several minutes (Case 32), and another, episodes of coughing which were followed by cyanosis and loss of consciousness (Case 36)

Muscular hyperactivity, spasms, hyperkinesias and the like frequently formed a part of the clinical picture, especially in later stages of the disease. Thus, there was muscular fibrillation, or twitching, in one region or another (Cases 18, 19, 22, 26 and 33), laryngeal spasms (Case 26), myoclonic contractions of eyelids, lips and upper limbs (Case 31), hiccup (Cases 6, 10, 18, 25 and 37), jerking automatic or purposeless movements (Cases 24 and 33), choreoathetotic movements of an upper limb (Case 37), tremor in one hand (Case 18), generalized tremulousness (Cases 15 and 32), horizontal nystagmus (Cases 18 and 21), and deviation of the head (Case 31) or eyes to one side (Cases 16, 20, 21, 24, 26 and 34)

Palsies in the realm of cranial nerves III, IV and VI were also observed anisocoria (7 cases), hippus (Cases 24 and 25), ptosis (Case 6), and strabismus (Cases 13, 22, 29 and 35) Partial bulbar palsy was noted in 4 cases (Cases 19, 22 and 36), and full-blown bulbar palsy associated with intercostal paralysis in 1 (Case 33)

Further clinical manifestations were positive grasp reflex (Cases 18, 21 and 31), opisthotonos (Case 6), muscular rigidity (Cases 22, 26 and 27) (which, in Case 22, was associated with trismus), vertigo (Case 7), pains in the limbs (Case 10) or neck (Case 32) and severe unilateral hyperpathia (Case 35) One

patient exhibited pronounced hyperglycemia (200 mg %) and mild glycosuria (Case 16).

Swelling of a cerebral hemisphere with shift across the midline was noted on ventriculography in 4 instances (Cases 4, 8, 18 and 37). The cerebrospinal fluid pressure was found increased in 13, reaching as high as 400 mm H_2O (Case 13), and was in the normal range (100 to 200 mm H_2O) in 5. Papilledema was prominent in 1 case (Case 35), suggestive in another (Case 2), and absent in 16. A cerebellomedullary pressure cone was noted post mortem in Case 8, a hippocampal cone in Case 18, and hippocampal and cingular in Case 15.

The spinal fluid was examined in all 38 cases. There was always pleocytosis. The number of cells per cmm. varied. They were less than 100 in 5 cases, between 100 and 500 in 12, from 500 to 1000 in 4, and reached 1000 to 2000 in 3. Lymphocytes prevailed, in about half the cases they constituted more than 90 per cent of the cells. Neutrophilic leukocytes predominated in one cell count or another in only 5 cases. Total proteins were above normal (45 mg per cent) in 28 of the 34 in which a determination was made, the value being as high as 320 mg per cent; they were in normal range in 5 cases, and rose from normal to an increased amount in 4. Erythrocytes were noted in the lumbar fluid in 5 cases in relatively early stages of the disease, and xanthochromia, later on, in 6. The gold curve was flat in 4 cases, was of the first zone type in 2 (Cases 26 and 30), of the second-zone in 1 (Case 34), and showed a slight rise in the first or second zone in 2.

Observation of the brain at autopsy usually revealed striking changes. Certain structures were softened and frequently hemorrhagic as well: temporal lobe (including the hippocampal formation and amygdala), insular region of the frontal lobe, anterior part of the gyrus cinguli, and the posterior orbital cortex (Figs. 9 and 18). The anterior part of the corpus callosum sometimes shared in the damage (Fig. 9). All or the majority of these structures were affected in 16 cases, unilaterally in about half, and bilaterally, though unequal in the two sides, in the other half. The temporal lobe was described as solely affected in 11, again



Figure 9 (Case 1, Table II, duration of illness, 5 days) Acute encephalitis of the herpes simplex type, showing hemorrhagic softening of the temporal lobes, posterior orbital gyri, and the region of the lateral cerebral fissure, including the insulae The gyrus cinguli and adjacent part of the corpus callosum are also affected (Courtesy of Dr O Stochdorph Dusseldorf, Germany)

either unilaterally or bilaterally. Softening also occurred in the occipital lobe (6 cases), parietal lobe (3 cases), "basal ganglia" (Case 19), thalamus (Cases 17 and 27), caudate nucleus (Cases 23 and 27), putamen (Case 14), globus pallidus (Case 17), lenticular nucleus together with the adjoining white matter (Cases 29 and 34), and the septal region and nucleus accumbens septi (Cases 14 and 31). Even more widespread softening of the brain of one or both sides was noted grossly in 3 instances the brain was said to be mushy in Case 2, semiliquid in Case 25, and gelatinous in Case 32.

On microscopic examination, the meninges frequently showed hyperemia, and, less often, accumulations of coagulated (edema) fluid in the arachnoidal meshes Meningeal cellular exudate, consisting of lymphocytes and plasma cells in vascular sheaths and predominantly of macrophages in archnoidal meshes, was pronounced in regions of cortical damage, and was frequently seen in limited amount over apparently unaffected cerebral cortex and around the cerebellum, brain stem, and spinal cord, as far down as lumbar segments (Case 24) Neutrophilic leukocytes were only occasionally encountered The number of cells in the meningeal exudate did not seem to change with time, but in cases of longer duration, macrophages were sometimes relatively more numerous The inflammatory reaction was not shared by the choroid plexuses, except for a few perivascular lymphocytes in the stroma in occasional cases.

A striking and constant feature in all the cases was necrosis of the temporal cortex accompanied by tissue breakdown The lesions were usually most pronounced subpially in upper laminae and perivascularly in middle and lower laminae (Fig 10A and 10B), with laminae III and V often the sites of predilection In numerous cases perivascular foci of rarefaction necrosis either dotted damaged regions indiscriminately (Fig 11A) or had a laminar distribution (Fig 7) In such foci, disintegration of nerve cells and macrophages was often in progress (Fig 11B), and it was within or adjacent to such foci that nuclear inclusions were often very frequent Cortical damage was not all inclusive, for occasional gyri in the midst of devastated regions were spared



Figure 10. *A* (Case 3, Table II; duration of illness, 7 days). Cerebral cortex, showing profound breakdown of the cortex with collection of vast numbers of mononuclear cells, many of them macrophages. The pia has been disrupted, allowing ingress of inflammatory cells into the subjacent cortex. A breakthrough of perivascular inflammatory cells into the cortical substance (in lower part of photograph) is also visible. X70. [illegible]) *B* (Case 12; duration of illness, 11 days). Upper and middle laminae of the cerebral cortex, showing profound inflammatory-cell and glial reaction around a vein. The perivascular space contains [illegible] cells, and in one region in particular (*arrow*), where the vessel wall is necrotic, many cells have broken through into the surrounding cortical substance. X100. *A and B* hematoxylin-eosin stain. *C* (Case 1; duration of illness, 5 days). Cerebral cortex, showing numerous fat-filled macrophages adjacent to the wall of a large vein (top of the [illegible]). X175. Oil red-O stain.

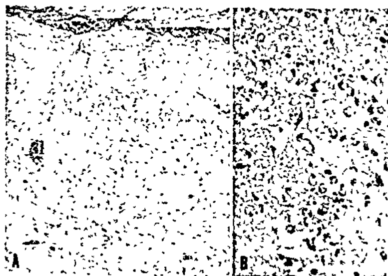


Figure 11 (Case 2, Table II duration of illness 6 days) *A* Foci of rarefaction necrosis in cerebral cortex Two of the foci are related to vessels, around one of which a hemorrhage has occurred Many of the nerve cells exhibit extreme pyknosis The meninges show numerous mononuclear cells, mostly lymphocytes X 35 *B* A focus of rarefaction necrosis in the subcallosal gyrus, containing macrophages some of which are disintegrating There were inclusions in numerous nerve cells adjacent to this focus X 300 Both stained by hematoxylin and eosin

The cortex at the depths of sulci frequently suffered somewhat more severely and, in some cases, more frequently than the cortex of the crests of gyri

Small hemorrhages, usually perivascular and often extending into the adjacent damaged nervous tissue, were very common In 5 cases the hemorrhages were relatively large

Sizeable hemorrhage was present in the putamen in *Case 9* (Table I), in the brachium conjunctivum and pars basilaris pontis in *Case 10* (Table II), in the putamen, amygdala and nucleus accumbens septi (from 2 x 2 mm to 18 x 10 mm) in *Case 14* (Table II) in the subcortical white matter and cortex, with break-through into the subarachnoid space in *Case 15* (Table II), and in the middle third of the pars basilaris pontis in *Case 32* (Table II)



Figure 14 (Case 7, Table II, duration of illness, 9 days) Reactive changes in the cerebral cortex. *A*. Proliferated cells in the subpial region A cell with similar features is to be noted in the subarachnoid space (*arrow*). X 450 *B* Cells highly similar to those in *A* are present perivascularly and diffusely These cells may represent both proliferated oligodendroglia and migrated ameboid cells X 165 Both stained by hematoxylin-eosin

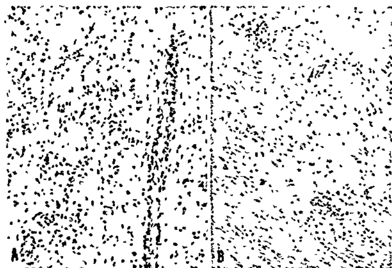


Figure 15 (Case 29, Table II, duration of illness, 17 days) *A*. Middle laminae of cerebral cortex, showing a few cells in the sheath of large and small vessels, enlarged astrocytic nuclei (*arrows*), and rod shaped cells Most of the nerve cells have disappeared X 100 *B*. Lower laminae of the cerebral cortex Cell nodules, apparently neuronophagic in character, and considerable loss of nerve cells There are only a few astrocytes and rod-shaped cells X 110. Both stained by hematoxylin-eosin

in regions of damaged temporal cortex as early as the 10th day (Case 9), and were seen in many of the cases from the 12th day onward

With the passage of time the pathological picture, in some cases at least, was more and more variegated Whether this was due to differing intensity of insult occurring simultaneously in different regions or to repeated recrudescence of insult could not be decided, though the latter seemed most likely

For example, in *Case 21*, of 14 days' duration, examination of different parts of affected cerebral cortex revealed the following patterns (1) Cortex extremely spongy with foci of rarefaction necrosis, many nerve cells and glia necrotic, practically no mesenchymal or glial reaction, (2) relatively longstanding foci of tissue breakdown with ameboid-cell reaction, and, especially in superficial laminae, dense astrocytosis, also many collec-



Figure 17 Perivascular changes in the cerebral white matter *A* (Case Table II, duration of illness, 17 days). Vessel in the subcortical wh matter, with small mononuclears, including ameboid cells, in its sheat Some ameboid and rod shaped cells are present in the surrounding whi matter Most of these are believed to have arisen from the vasculr sheat rather than from Hortega microglia X 305 *B* (Case 2, duration of illnes 6 days) Subcortical white matter Lymphocytes prevail about much o the vessel, except at one end, where, in the region of invasion of the whit matter, the cells are decidedly pleomorphic (ameboid histiocytes). X 305 *C* (Case 31, duration of illness, 25 days) Subcortical white matter Large perivascular collections of mononuclear cells with invasion of surroundings and reaction on the part of glia Similar foci were present somewhat deeper in the white matter X 150 All stained by hematoxylin-eosin

parietal region (Cases 1 and 31) Case 16 was of interest in that white matter beneath severely damaged cortex had broken down into cystic cavities, but only two blocks from the brain were available and the region of the cortex from which they were taken was unkown In occasional cases of longer duration (e g , Case 21)

astrocytic nuclei were enlarged, numerous fibrillary astrocytes, positive in Holzer crystal-violet-stained preparations, had formed at considerable distances deep in the white matter. Only rarely in our cases (e g, Case 2) was there significant involvement of the white matter in regions where the cerebral cortex was relatively spared

Other regions of the brain were significantly affected, but not as consistently as the temporal cortex. The part of the brain which stood out because of the frequency and severity with which it was damaged was that at the level of the anterior perforated substance The structures affected, though in various combinations, were the ventral part of the septum pellucidum, subcallosal and parolfactory gyri, anterior commissure (median part), nucleus accumbens septi ("olfactory striatum"), sublenticular or subpallidal tissue, anterior perforated substance, and amygdala In all 16 cases in which representative sections from these structures were available for study, the lesions were striking and consisted, variously, of perivascular damage of tissue with invasion by ameboid cells from neighboring vascular sheaths, more or less widespread nerve-cell necrosis, patchy tissue breakdown and, from about the 12th day onward, astrocytosis and appearance of rod-shaped cells The anterior perforated substance was damaged in 12 of the 16 cases, mostly in its lateral part, and the amygdala in 11, with the most pronounced change dorsally As a rule, these structures were affected bilaterally, but in different degree

The *thalamus* came next in frequency of involvement, though perivascular inflammatory-cell changes in the subependymal tissue were common The changes in the thalamus, of the same general nature as those just recounted, were relatively slight or moderate in half the cases, and severe in the other half The medial nuclei and pulvinar were the sites of predilection The *hypothalamus* was usually affected in moderate degree, with the chief changes consisting of inflammatory cells perivascularly, occasional breakdown of perivascular tissue, and occasional neuronophagic or gliomesenchymal nodules There were no special sites of predilection, though occasionally the lesions were most prominent in the dorsal part of the hypothalamus Formation of gliomesen-


A


B

chymal cell nodules and diffuse invasion of the tissue by ameboid cells were seen in the supraoptic nucleus and neurohypophysis in one instance (Case 15) (Fig. 19). Along the ventricular surface of the *head of the caudate nucleus*, perivascular lymphocytes were fairly common, but the nucleus itself was severely affected in only 2 cases, slightly affected in about half the cases, and not at all in 6 The *putamen* was somewhat more often damaged, usually laterally, in relation to necrotic insula and claustrum, or ventrally, near damaged sublenticular tissue. As to the *globus pallidus*, it was often spared, and when involved, usually showed acute nerve-cell damage In 5 cases it was severely affected, usually by contiguity from damaged sublenticular tissue. The *subthalamus* contained lesions in only occasional cases, and the *geniculate bodies*, too, the medial more often than the lateral Thus, the lesions decreased significantly in the direction of the midbrain

On the whole, the *brain stem* was comparatively little affected except for the pars basilaris pontis There was no apparent increase in the severity of the lesions in the brain stem with time Nerve cells, in general, showed little evidence of regressive changes The *midbrain* was spared in about one-third of the cases Often there was little more than scattered perivascular collections of lymphocytes and ameboid cells, with the latter now and then

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Figure 18 (Case 10, Table II duration of illness, 10 days) *A* Hemisphere stained by cresyl echt violet, showing necrosis of practically all the temporal cortex and widespread damage of the white matter, as manifested by an opaque quality Many of the vessels are surrounded by inflammatory cells. Other parts of the cortex are also affected the posterior orbital cortex the gyrus cinguli (and subjacent white matter), the adjacent medial frontal gyrus, the middle frontal gyrus (where there are multiple foci of rarefaction necrosis), the inferior frontal gyrus (adjacent to temporal cortex), and the nucleus accumbens septi (best seen in *B*) *B* Profound demyelination of the temporal lobe and posterior orbital gyri Slight demyelination is to be noted in the inferior frontal gyrus (near the mouth of the sylvian fissure) and in the gyrus cinguli Weil myelin method

invading damaged nervous tissue around the vessels. Small glio-mesenchymal cell nodules were fairly common in some cases, while neuronophagic nodules were few. Enlarged astrocytic nuclei were commonly present in cases of longer duration Rod cells

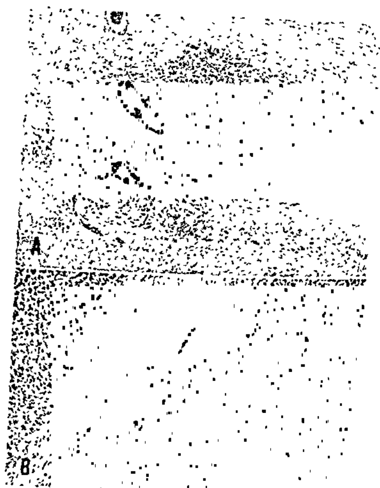


Figure 19 (Case 15, Table II, duration of illness, 12 days) *A* Supraoptic nucleus, showing intense perivascular inflammatory reaction with myriad mononuclear cells in the interstitium X 42 *B* Pituitary gland, showing large mononuclear cell accumulations, with tissue necrosis in the posterior lobe To the left are the anterior lobe and infundibular stem X 80 Both stained by hematoxylin-eosin

in the region of tissue damage were always few, but seemed more numerous in cases of longer duration. The distribution of the lesions was not consistent. They were in periaqueductal grey, tegmentum (usually in the region of the median raphé) and substantia nigra alike, though, as a rule, they predominated in the ventral tegmentum and substantia nigra.

On the other hand, the *pons* was consistently affected Here, lymphocytes and plasma cells were found perivascularly in the floor of the IVth ventricle and in the region of the median raphé, and, in the tegmentum, scattered collections of ameboid cells and activated glia in perivascular nervous tissue, and gliomesenchymal cell nodules. The nucleus loci caerulei and environs were often strikingly affected Lesions in the pars basilaris were of the same nature as in the tegmentum, but were almost always more intense

The *medulla oblongata* seldom showed more than perivascular collections of lymphocytes and plasma cells in the region of the IVth ventricle and median raphé, and, in the grey matter at the level of the pyramidal decussation, scattered gliomesenchymal cell nodules as well The inferior olivary nucleus was seldom affected In one case characterized clinically by bulbar palsy (Case 33), insufficient material was available to determine its cause.

In the *spinal cord,* sparse lesions of the same nature as those in the brain stem were found in the grey matter in 4 of the 9 cases in which sections of spinal cord were available for study (Cases 3, 14, 20 and 25), *and in one the lesions involved the pericornual* white matter as well (Case 20).

Examination of the *cerebellum* in 13 cases revealed changes in only 4—in Case 12; damage of the Purkinje and molecular layers with reactive changes, and with neuronophagic nodules and proliferated glia in the dentate nucleus, and in Cases 3, 14 and 20, perivascular collections of inflammatory cells and reactive cells in the surrounding white matter

Varying numbers of *cranial nerves* (III-XII) were examined in 7 cases Only in Cases 6, 7 and 20 were some of them affected, and then in the form of a slight interstitial neuritis and, here and there, segmental neuritis (of Gombault Stransky) (Case 20); in Case 20 the gasserian ganglion showed dense collections of in-

flammatory cells in the dura, moderate proliferation of Schwann cells, occasional macrophages within Schwann tubules, and slight hyperplasia of capsular cells. Olfactory bulbs and tracts were available for study in 5 cases (Cases 2, 5, 6, 20 and 21). In all, there were occasional, more or less conspicuous collections of perivascular lymphoid cells, perivascular invasive cells, a few scattered small cell-nodules, and occasional rod cells. The lesions were of the same intensity on the two sides in half the cases, and were somewhat more pronounced unilaterally in the others. The spinal roots showed little of note except for posterior-root ganglionitis in one case (Fig. 20).



Figure 20. (Case 20, *Table II, duration of illness 13 days.*) Posterior cervical root ganglion, showing numerous mononuclear cells in the interstitium, loss of ganglion cells, and proliferation of capsular cells. X 120. Hematoxylin-eosin stain.

7. SUBACUTE ENCEPHALITIS WITH INCLUSIONS

Our chief clinical criteria of "subacute inclusion encephalitis" —as the disorder is usually called—were gradual onset of illness without fever and an ingravescent course There were 7 cases in this group, most of which have been previously published (Table III). Virus isolation was attempted only in Cases 1 and 2, with negative results; in Case 1 extensive studies were carried out,* and in Case 2, brain material was introduced intracerebrally into mice and injected into embryonated eggs

Duration of illness ran from 4 months to 7 years Ages of the patients ranged from 5 to 17 years except in Case 7, which is discussed separately at the end of this Section In the 6 cases in children and adolescents the clinical picture was most diverse Mental symptoms were among the earliest to appear, and they progressed rapidly to a state of dementia. One patient was committed to a mental institution (Case 6), and only in this case were remissions observed Among the clinical disturbances were hallucinations (Cases 1 and 3), aphasia (Cases 2 and 5), seizures of various kinds (Cases 2, 3, 4, 5 and 6), "startle attacks" (Case 2), myoclonic spasms or movements (Cases 1, 3, 4 and 5), hyperkinesias (Cases 2, 3 and 5), muscular rigidity (Cases 1 and 2), and signs of cranial-nerve involvement (Cases 3, 4 and 5) Optic atrophy was also encountered (Cases 4 and 6). Fever occurred during the course of illness in half the cases (Cases 2, 3 and 5)

Thus, the clinical manifestations in our cases were most diverse. Cases reported by others as well as by one of us (L v B) have often been characterized, for weeks on end, by a periodic stereotyped rhythmic jerking of one or more limbs or by attacks of loss of muscular tonus ******

Gross softening was described as occurring in the base of the temporal and frontal lobes in Case 3, the right temporal pole in Case 4, and most of the cerebrum bilaterally in Case 7 In Case 2 there was conspicuous softening not only of the left prefrontal region, but also, though in smaller degree, of all lobes bilaterally Elsewhere in these cases the brain was usually firmer than normal Only in 1 of the 37 cases of subacute sclerosing leukoencephalitis studied by one of us (L v B) was cortical softening detected,

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Figure 20 (Case 20 Table II, duration of illness, 18 days) Posterior cervical-root ganglion, showing numerous mononuclear cells in the interstitium, loss of ganglion cells, and proliferation of capsular cells X 120 Hematoxylin-eosin stain

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Figure 20. (Case 20, Table II, duration of illness, 13 days) Posterior cervical root ganglion, showing numerous mononuclear cells in the interstitium, loss of ganglion cells, and proliferation of capsular cells. × 120. Hematoxylin-eosin stain.

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Gross softening was described as occurring in the base of the temporal and frontal lobes in Case 3, the right temporal pole in Case 4, and most of the cerebrum bilaterally in Case 7 In Case 2 there was conspicuous softening not only of the left prefrontal region, but also, though in smaller degree, of all lobes bilaterally Elsewhere in these cases the brain was usually firmer than normal Only in 1 of the 37 cases of subacute sclerosing leukoencephalitis studied by one of us (L v B.) was cortical softening detected,

TABLE III

CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness	Clinical Features	Spinal Fluid Protein (mg %)	Cells (cmm) (% lymphocytes) Other Data	Pertinent CNS Changes
1 796868 Case 2 of Dawson[a]	5 yr F	4 mo	Onset awoke at night laughing & crying alternately, had hallucinations Later jerking mvts of limbs which became violent & more frequent (occas so rapid that patient trembled all over), mute, stuporous, musc rigidity, temp 107 4°F	incr — CSF pressure slightly increased or normal	2 4	*Gross* Petechiae in meninges, cerebral cortex, white matter & basal ganglia *Microscopic Cerebral cortex*, focal nerve-cell loss, scattered neuronophagic nodules, many rod cells, mod astrocyte response, sparse perivasc lymphocytes & plasma cells (Fig 22A); hippocampus, mod affected *Cerebral white matter*, slt. astrocytosis subcortically. *Caudate & lentic. nuclei*, slt nerve-cell loss, a few rod cells, occas neuronophagic nodules; *pons*, occas perivasc. lymphocytes, slt. nerve-cell loss, occas patchy astrocytosis, *med. obl*, occas neuronophagic nodules in inf. oliv. nuc with many rod cells *Spinal cord*, occas perivasc. lymphocytes & plasma cells *Cerebellum*, no change

TABLE III—(Continued)
CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness	Clinical Features	Spinal Fluid Protein (mg %)	Spinal Fluid Cells (cmm) (% lymphocytes)	Pertinent CNS Changes
2 706862 WU 12870 Smith et al*	16 yr M	7 mo	Early complaint "black spots before eyes" 2 wk later when startled had attacks (once a wk for 10-15 sec then daily) of dizziness, staggering & falling backward 4th mo aphasic, numerous "spastic attacks," 5th mo musc rigidity, periodic grimaces & carpopedal like spasms Later mute, limbs fixed in position of carpopedal spasm, frequent convulsive seizures, fever begun at 6th mo (to 101°F)	18 26 27 Other Data: CSF pressure 210 mm H_2O Gold 0000000000 1111100000	1 1 2	*Gross* Slt cortical & subcortical softening, lt. precentral gyrus, same but less pronounced in frontal, parietal, temp & occip regions, bilat *Microscopic* *Lt. precentral gyrus*, end stage of necrosis with abundant astrocyte & macrophage response & many perivasc inflam cells (Figs 21A & 21B) *Cortex in general*, much nerve cell loss, advanced astrocytosis & frequent neuronophagic nodules, a few rod cells & sparse perivasc inflam cells, *except for lt precentral gyrus*, changes most pronounced in cingulate & occip. cortex, hippocampal formation, many inclusions, with bilat nerve-cell loss & demyelination (Fig 21C) *Cerebral white matter*, demyelination in region of lt cingulate & sup. frontal gyri (Fig 21C), relatively little involvement elsewhere, though intense sclerosis occas present beneath intact cortex *Lt parolfactory area*, slt gliosis *Basal ganglia*, no change *Brain stem*, regressive changes in nerve cells, sparse perivasc inflam cells *Cerebellum & spinal cord*, no change Many nuclear inclusions (Figs 1B & 1C), a few in cytoplasm

TABLE III—(Continued)

CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No. AFIP No. Other Nos	Age at Onset Sex	Duration of Illness	Clinical Features	Spinal Fluid: Protein (mg.-%)	Cells (cmm.) (% lymphocytes)	Pertinent CNS Changes
				Other Data		
3 197269 Malamud, Haymaker & Pinkerton*	17 yr M	9 mo	Failure to recognize objects & people, laughter without cause 4th mo. (in hosp.) auditory hallucinations, delusions musc. twitching & myoclonic jerks of limbs & eyes, waxy flexibility, pill-rolling tremor, ptosis of lt. eyelid, fixation of rt. eye on attempted accom.-converg., one convulsion. Fever at 5th mo. (100-105°F)	62 18 18-43 No papilledema CSF pressures "normal", 165 mm H_2O Gold. 5555543210	5 (100) 6 (100) 0.6	Gross. Base of temp. & frontal lobes softened & had gelatinous appearance; parietal, occip., lat. temp. & frontal lobes firm with gyri small. *Microscopic*. *Cerebral cortex*, many dense perivasc. cell-collections, widespread nerve cell loss, rod cells in all laminae, hyperplastic astrocytes in deeper laminae. *Cerebral white matter*, widespread involvement, with gliosis more pronounced than demyelination, many fat-containing macrophages, with some in adjacent cortex (Fig. 25A). *Thal. & lat. genic. body*, changes similar to those in cerebral cortex; *caudate nuc., putamen, glob. pall. & hypothal.*, same types of lesion, but less pronounced. *Midbrain*, involved only in periaqueductal grey (perivasc. lymphocytes); *pons*, pars basilaris severely affected, with marked nerve-cell loss, advanced astrogliosis & massive perivasc. collections of lymphocytes & plasma cells (Fig. 25C). brachium pontis, less affected. *med. obl.*, scattered cell-infiltrates dorsally. *Cerebellum*, occas. perivasc. cell infiltrates in white matter. *Spinal cord*, no change.

TABLE III—(Continued)

CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness	Clinical Features	Spinal Fluid Protein (mg %)	Cells (cmm) (% lympho-cytes)	Pertinent CNS Changes
				Other Data		
4 201977 Malamud Haymaker & Pinkerton[99]	9 yr F	21 mo	1st few days irritable fell frequently lost control of lt hand 4th mo (in hosp) vomited continually, transient opisthotonos 8th mo impaired speech clonic spasms of face & limbs 10th mo dysphagia frequent convulsions severe optic atrophy high fever	25	0	*Gross* Softening of rt temp pole, rest of brain, firm *Microscopic Cerebral cortex*, all laminae affected in several regions numerous perivasc cells, striking nerve cell loss, diffuse sclerosis & many rod cells & hyperplastic astrocytes (Fig 25B) *Cerebral white matter*, widespread gliosis & some demyelination, perivasc macrophages *Caudate nuc, putamen & thal*, much nerve cell loss, dense sclerosis, hyperplastic astrocytes, minimal inflam-cell reaction *Brain stem*, severely involved (with many microglial nodules), espec tegm & pars basilaris pontis & dorsal & olivary parts of med obl *Cerebellum* diffuse degen of Purkinje cells & cells of dentate nuc with slt involvement of white matter *Spinal cord*, in grey matter & in less degree in surrounding white matter perivasc inflam cells, advanced gliosis & occas glial nodules (Fig 26)
				Increfsed intracranial pressure,' then 175 mm H_2O Ventriculogram no brain shift		

TABLE III—(Continued)

CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AHP No Other Nos	*Age at Onset Sex*	*Duration of Illness*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
5 WU 17535 Landau & Gitt[14]	10 yr M	6½ yr	For 2 wk had headaches, in following months, mental deterioration 1st 3 mo of present illness daily loss of consciousness Later (in hosp) temp 99.8°F, diplopia, musc incoordination 5th mo polyphagia, transient unconsciousness, attacks of musc jerking (with ballistic mvts) Later temp to 101.5°F, lethargy, homonymous hemianopia, jargon aphasia	51 30 "normal"	6 (0) 1 "normal"	*Gross.* No data available *Microscopic:* Biopsy (1 yr. after onset). *Meninges,* lymphocytes, plasma cells & activated histiocytes *Cerebral cortex,* perivasc. lymphocytes & plasma cells, nerve cell degen., proliferation of microglia & astrocytes, glial nodules *Subcortical white matter,* rarefaction, gliosis (astrocytes & microglia), small retracted glial scar. No inclusions in biopsy material, but many found post mortem, both nuclear & cytoplasmic (Fig. 1A) (Report being prepared by Luce, Landau & Smith)
				No papilledema CSF pressure 170 mm H_2O		

TABLE III—(Continued)
CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AFIP No Other Nos	Age at Onset Sex	Duration of Illness	Clinical Features	Spinal Fluid Protein (mg %)	Spinal Fluid Cells (cmm) (% lympho-cytes)	Other Data	Pertinent CNS Changes
6 218126 LPC-393 Malamud Haymaker & Pinkerton[80]	10 yr F	7 yr	First 6 wk attacks of staring expression, tonic musc contraction & falling to ground, impaired vision of lt eye 7th wk (in hosp) temp normal, intellectual deficit, slt rt optic atrophy, bilat degen of macula 4th mo mute, attacks of unconsciousness 3d yr dementia 6th yr blindness, jacksonian like attacks in rt lower limb 3 times weekly, spasticity Periods of partial remission during course	—	—	—	*Gross.* Meninges diffusely thickened, mod gyral atrophy White matter of frontal lobe, grey & gelatinous, in other lobes unusually white & firm. *Microscopic Cerebral cortex,* widespread involvement with variable nerve cell loss, *in some regions,* small gliomesenchymal cell nodules & scattered rod cells; *in others,* involvement of most laminae by perivasc inflam. cells, small cell nodules (neuronophagic & perivasc), microgliosis & astrocytosis (in deeper laminae); *in still other regions,* severe nerve-cell loss & gliosis & sclerosis of all laminae *Occip. lobe* most affected, *cingulate gyrus,* little *Cerebral white matter,* advanced gliosis & some demyelination (Fig 24). *Severely involved* thalamus, with many perivasc cells, glial nodules, striking nerve cell loss, intense astrocytosis & microgliosis (Fig 25B) *Mod involved* hippocampal formation (Fig 23A), caudate nuc, putamen, genic bodies, pretectal region, subst nigra, tectum & tegmentum, pontile (pars basilaris) & dentate nuclei, & med obl (floor of IVth ventr, retic form, & inf oliv nuc) *Slight* glob pall, hypothalamus, red nuc, & Purkinje cells *Spinal cord,* not available

TABLE III—(Continued)

CASES OF INSIDIOUS ONSET SUBACUTE INCLUSION ENCEPHALITIS (7 CASES)

Case No AFIP No Other Nos	*Age at Onset Sex*	*Duration of Illness*	*Clinical Features*	*Spinal Fluid Protein (mg %)*	*Cells (cmm) (% lymphocytes)*	*Pertinent CNS Changes*
				Other Data		
7 265125	36 yr M Negro	'Months'	For "months" had periodic stiffness of legs, jerking of lt leg & abdom cramps, and in final 2 wks headaches, nausea & occas vomiting 1st day of acute illness (12 days' duration) severe headache, slt fever, jerking mvts of lt leg, widespread trembling 2d day, went to work 3d day, found wandering through streets aimlessly Later (in hosp) Combative, euphoric, stiff neck & back, spastic, f convulsions, temp 104.2°F	21 18 CSF pressures 280 & 300 mm H_2O Gold 0000000000 1221000000 No papilledema (2 exam s)	73 37 (100) 17 (100)	*Gross* Brain of pink hue & very soft throughout, stuck to knife when sectioned *Microscopic* Pathological picture highly similar to that in previous cases *Cerebral cortex*, in some regions, profound nerve cell loss, advanced astrogliosis & abundant rod-cell formation (Fig 22B); scattered perivasc. accumulations of lymphocytes & plasma cells (in meninges too), some gyri very sclerotic *Cerebral white matter*, slt astrocytosis *Striatum, pallidum, thalamus, pons & cerebellum*, spared



Figure 21 (Case 2, Table III duration of illness 7 months) Subacute inclusion encephalitis *A* Left precentral gyrus which was found softened on gross examination The destructive process involves much of the cortex and the subjacent white matter *B* Enlargement of a field from the lower cortex in *A*, showing macrophages and other mononuclear cells perivascularly, and complete replacement of the cortical substance by abundant plump astrocytes, macrophages, and other cells ×90 Hematoxylin-eosin stain C A more caudal level on the left side. Demyelination is fairly prominent in the hippocampal formation and gyrus cinguli, and slight in the medial superior frontal gyri Pal Weigert myelin stain

and it was exceedingly minor "

Microscopically, the meninges contained rather few collections of lymphocytes, plasma cells, and activated histiocytes As a rule, the reaction was focal, being limited for instance, to the meninges at the base of a sulcus

Regions of cerebral cortex observed grossly to be softened usually had a sclerotic appearance microscopically. In one instance (Case 2, Table III) the softening was characterized by rather



Figure 22 Subacute inclusion encephalitis *A* (Case 1, Table III; duration of illness 4 months) Middle laminae of the cerebral cortex. A few nerve cells have disappeared, perivascular spaces contain a slight excess of inflammatory cells rod cells have accumulated, and there is one neuronophagic nodule *(arrow)* (The overlying meninges contained rather sparse lymphocytes and macrophages) X 120 (Courtesy of Dr John L. Shapiro Nashville Tenn) *B* (Case 7, duration of illness, "several months") Middle laminae of the cerebral cortex Practically all the nerve cells have disappeared and there are abundant plump astrocytes and rod cells X 195 Both stained by hematoxylin-eosin

circumscribed destruction of the cortex and the underlying white matter, with pronounced gliomesenchymal reaction (Fig 21). In general, the changes in the cerebral cortex were rather wide-

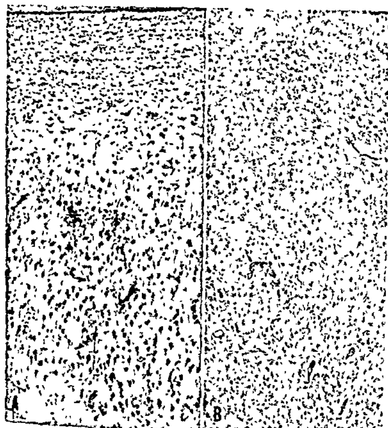


Figure 23 Subacute inclusion encephalitis *A* (Case 6, Table III, duration of illness, 7 years). Pyramidal layer of the hippocampus, showing neuronophagic nodules, an abundance of rod cells, and a few perivascular mononuclear cells X 110 Cresyl echt violet stain *B* (Case 4, duration, 21 months). The cerebral cortex is extremely sclerotic, and contains many fibrillary astrocytes and rod cells Practically all the nerve cells have disappeared Occasional vessels contain a few mononuclear cells in their sheaths X 48 Hematoxylin-eosin stain (From Malamud Haymaker and Pinkerton [60])

only in regions of pronounced cortical involvement, i e , it was of a focal nature, as in many of the acute cases of encephalitis A case of this kind is illustrated in Figure 22. In the 3 other cases (Cases 3, 4 and 6), however, damage of the cerebral white matter was widespread, pronounced demyelination and gliosis occurring *even in the regions where cortical involvement was minimal* (Fig 24). Fat filled macrophages were in abundance in affected white matter (Fig. 25A)

A pathological process of much the same type occurred in other parts of the central nervous system, but not as consistently



Figure 24 (Case 6, Table III, duration of illness 7 years) Alterations in the white matter in subacute inclusion encephalitis *A*, stained by the Weil method, shows demyelination subcortically, and *B*, stained by the Holzer method, pronounced gliosis of both subcortical and intragyral white matter (The same changes were widespread in the cerebrum) X 6



Figure 25 Subacute inclusion encephalitis *A* (Case 3, Table III, duration of illness, 9 months) Cerebral cortex and white matter stained by scarlet red (with hematoxylin as a counterstain), showing an abundance of fat filled macrophages in the white matter and, in the cortex, neuronophagic nodules, rod cells, and other elements X 100 *B* (Case 6 duration of illness 7 years) Thalamus, illustrating huge perivascular cuffs of small mononuclear cells and gliomesenchymal cell nodules X 100 Cresyl echt violet stain *C* (Case 3, duration of illness, 9 months). Pars basilaris pontis, showing porosity and gliosis of the nuclei pontis and perivascular collections of mononuclear cells The pyramidal bundle (to the right) is spared X 36 Hematoxylin-eosin stain

as in the cerebral cortex and white matter. The thalamus (Fig 25B) was a site of predilection, but it was moderately or severely involved only in Cases 3, 4 and 6 Less commonly and less severely affected were the caudate nucleus and putamen. In the 6 cases in which the pons was available for study, the pars basilaris was severely affected in 3 (Cases 3, 4 and 6), slightly in 2 (Cases 1 and 2), and was spared in 1 (Case 7). Occasionally the tegmentum of the pons shared prominently in the pathological process, as did also the geniculate bodies Only minor changes were found in the globus pallidus, hypothalamus, and medulla oblongata In the 4 cases in which sections of spinal cord were available, the grey matter was slightly affected in 1 (Case 1) and markedly in the other, with the lesions extending for a distance into the adjoining white matter (Fig 26). The cerebellum was spared in



Figure 26 (Case 4 Table III duration of illness 21 months) Subacute inclusion encephalomyelitis Spinal cord at upper cervical level showing profound nerve-cell loss, increase in glia, and accumulation of mononuclear cells perivascularly Both anterior and posterior horns are affected *B* illustrates a higher magnification of the posterior horn and adjacent white matter The grey matter showing striking increase in glia presents a sclerotic appearance Reaction in the white matter is much less pronounced *A*, X 23 *B*, X 62 Hematoxylin-eosin

Cases 1, 2 and 7, but in Case 3 the white matter showed occasional inflammatory-cell infiltrates, in Case 4, diffuse degeneration of the Purkinje-cell layer and dentate nucleus, and in Case 6, patchy degeneration of the Purkinje-cell layer and dentate nucleus Data on the cerebellum in Case 5 are not available.

Case 7 is treated separately because the patient was an adult and because of the paucity of symptoms prior to the acute illness, which lasted 12 days The cerebral cortex presented a pathological picture characteristic of subacute inclusion encephalitis (Fig 22B). The pathological process had obviously been going on for a period longer than 12 days.

8. DISCUSSION

Various forms of viral disease associated with nuclear inclusions have been dealt with in the foregoing pages, but we are concerned in this discussion only with the acute and subacute forms of inclusion encephalitis Our series of 53 cases, of which 17 have been previously published, were divided arbitrarily into three groups (Tables I, II and III) in order to determine more readily whether or not clinical and pathological differences existed between them. Age incidence varied in the different groups, the onset occurring in all decades in the acute type, with a peak in individuals in their 20's, while in subacute inclusion encephalitis the disease was limited to children and adolescents, except in one instance in which the patient was an adult (Table IV). In our series, no special seasonal incidence was found (Table V) The proportion of males to females affected was 24:19.

The 2 cases in which the infection was acquired at the time of birth or shortly afterward stood apart pathologically from the others in that the major changes were in the viscera, especially the liver and adrenal cortex, while the brain was only minimally affected On the other hand, in infants 8 days of age or older the lesions in the central nervous system were no different from those in adults, except that the basal ganglia and brain stem were sometimes the seat of more severe and widespread lesions. The cerebral cortex was widely involved in some of the infants, but this

TABLE IV

AGE AT ONSET OF ACUTE AND SUBACUTE INCLUSION ENCEPHALITIS IN 53 CASES

Groups	*0-10 days*	*11-30 days*	*2-6 mo*	*7-12 mo*	*2-9 yr*	*10-19 yr*	*20-29 yr*	*30-39 yr*	*40-49 yr*	*50-59 yr*	*60-69 yr*	*Unknown*	*Total*
Acute encephalitis in infants (Group 1)	3	3	1	2									9
Acute encephalitis in adolescents and adults (Group 2)						2	15	5	5	6	2	2	37
Subacute encephalitis (Group 3)					2	4		1					7

TABLE V

MONTHLY INCIDENCE OF ACUTE AND SUBACUTE INCLUSION ENCEPHALITIS IN 53 CASES

Groups	*Jan*	*Feb*	*March*	*April*	*May*	*June*	*July*	*Aug.*	*Sept*	*Oct.*	*Nov*	*Dec.*	*Unknown*	*Total*
Acute encephalitis in infants (Group 1)			1	1		1	1		2		1	2		9
Acute encephalitis in adolescents and adults (Group 2)	5	2	1	2	1	1	6	3	2	3	2	4	1	37
Subacute encephalitis (Group 3)										2			5	7

was true also in some adolescents and adults; for example, in 3 adults an entire hemisphere or the whole brain was described on gross examination as mushy, gelatinous, or semiliquid (Cases 3, 25 and 32; Table II).

Most of the cases of encephalitis with an acute onset fell naturally into a group because of the character and topography of the lesions The necrosis and the distribution of the lesions in the cerebral cortex were features which, to our knowledge, are not duplicated under any other pathological condition. The picture was, indeed, so distinctive as to allow the prediction of the presence of nuclear inclusions of the herpetic type before they were found. In our entire series of acute cases, there was only one exception to the rule that profound damage occurs, and that was in a case in which the cortex showed only slight perivascular tissue destruction (Case 5, Table I).

In classifying our cases we had difficulty in deciding where to list some of them, whether with the acute cases or with the subacute. In Case 36, for example, of 3½ months' duration, there was some question whether the increasing forgetfulness and loss of weight over some months represented an early phase of the disease or were coincidental Since acuteness of the onset of illness was chosen as the criterion for including this and the other cases in Table II, we included Case 36 in Table II The same criterion led us to include Case 37 in Table II even though the patient, tided over, *probably, by decompressive craniotomy has survived* his encephalitis 4 years The same problem presented itself in regard to Case 7 in Table III Because of the acuteness of onset, this case was originally included in Table II, but when, on microscopic examination, it was apparent that we were dealing with subacute inclusion encephalitis (Fig 22B), the case was transferred to Table III There had been periodic spontaneous jerking movements of the limbs over several months, which were considered to have marked the onset Such cases raise the question whether the infection may not, for a time, be of a smoldering nature, and then break out in full flame. Equally vague in onset, suggesting smoldering infection, were Cases 4, 8, 15, 23 and 33 (Table II).

Direct evidence that the subacute form is a smoldering form of herpes simplex or other infection is lacking, for, in virus studies, agents have either not been isolated,[illegible] or, when isolated, have not been identified or established as the cause of the disease state.[illegible] In a virological study of one case reported by Foley and Williams,[illegible] Hurst isolated an agent which had all the characteristic features of the herpes simplex virus, but he reached the conclusion that it was not the cause of the subacute encephalitis because the patient was sero-negative for herpes simplex (as were also 3 other patients) and because in other similar cases he was unable to isolate an agent. In rabbits, either acute or subacute encephalitis may occur following introduction of the herpes simplex virus into the cornea.[illegible] According to Doerr and Zdansky,[illegible] most rabbits die from encephalitis within 2 days after inoculation. Others that survive the acute phase either grow better or worse with time. In one rabbit strain, a 30-day latent period followed inoculation, after which the illness became progressively severe and lasted as long as 36 days. If in rabbits of this strain the virulence of the virus was increased through animal passage, the latent period became progressively shorter, and death from encephalitis occurred in as short a period as 3 days.[illegible] Others, too, have shown that rabbits respond differently to the inoculation of herpes simplex virus, some die rapidly with acute encephalitis, others have an ingravescent illness, while still others are asymptomatic until encephalitis is set into being through anaphylactic shock.[illegible]

Before comparing the acute and subacute forms of the encephalitis further, let us inquire into the relationship which subacute inclusion encephalitis bears to subacute sclerosing leukoencephalitis. The latter is characterized by a slowly progressive illness in which the clinical picture is highly similar to that of subacute inclusion encephalitis. Early intellectual deterioration is followed rapidly by loss of all psychic function until dementia occurs. Involuntary motor activity develops, as does also progressively more pronounced muscular hypertonia consistent with increasing decortication. A low-grade fever frequently occurs.[illegible]

[illegible] The illness usually runs a course of 3 to 6 months, but may last a year or two.[illegible] With rare exceptions (an adult aged 34 without post-mortem verification[illegible] and our Case 7, Table III) only children and adolescents (as old as 20 years[illegible]) contract the disease

The structures chiefly affected in subacute sclerosing leukoencephalitis are the cerebral white matter, cerebral cortex, and thalamus Less affected than these are the subthalamic region, pars basilaris pontis, inferior olivary nucleus, and spinal cord, and least affected are the striatum, pallidum and the dorsal part of the brain stem In subacute inclusion encephalitis the predominantly affected structures listed by Greenfield[illegible] were occipital cortex and white matter, caudal part of the thalamus, pars basilaris pontis, and inferior olivary nucleus In dealing with the pathological changes in both subacute forms, Greenfield, in an *article by Foley and Williams,*[illegible] *summarized the changes as follows.* The occipital and temporal cortex were more affected than the frontal and hippocampal (though Sommer's sector was particularly involved), the basal ganglia showed subacute changes—with the thalamus most heavily affected and the caudate and lenticular nuclei slightly—the pontile nuclei were prominently involved and the midbrain moderately, the spinal cord was sometimes affected, and *the cerebellum was spared Thus, in the main,* the topography of the lesions in the grey matter in subacute inclusion encephalitis and subacute sclerosing leukoencephalitis are highly similar.

The cellular reactions in these two conditions are also basically the same Thus, in a representative case of subacute sclerosing leukoencephalitis (which was previously published[illegible]), there was profound gliosis of parts of the cerebral cortex and widespread and severe demyelination and gliosis of the white matter with macrophage response (Figs. 27 and 28), with the changes in both grey and white matter closely simulating those in a case diagnosed as subacute inclusion encephalitis (Figs 22B and 24) Nuclear inclusions were found in this case Another case of subacute sclerosing leukoencephalitis which may be cited is one in which the



Figure 27 Subacute sclerosing leukoencephalitis Cerebral hemisphere at the level of the uncus hippocampi, showing profound demyelination The myelin loss is most pronounced in the temporal lobe and gyrus cinguli, and is striking also in the centrum semiovale. Demyelination is also visible in the corpus callosum Pal-Weigert stain IB No 57/53, AFIP Acc 795110

cerebral cortex was moderately affected (Fig 29A) and showed numerous nuclear inclusions (Fig. 29B and 29C), and the white matter severely demyelinated, and, in some regions, completely broken down (Fig 29D) It should be mentioned that other authors, too, have observed inclusions in subacute sclerosing leukoencephalitis [illegible]

In agreement with others, [illegible] we would conclude, therefore, that subacute inclusion encephalitis and subacute sclerosing



Figure 28 Same case as in Figure 27 *A* Cerebral cortex, showing myriad plump astrocytes and a few mononuclear cells perivascularly and in the meninges Practically all the nerve cells have disappeared X 195 Hematoxylin eosin stain *B* Hyperplastic fibrillary astrocytes in the subcortical white matter and a few in the lowermost laminae of the cortex X 145 Holzer stain C. Cerebral white matter, stained by Scharlach R hemalum, showing numerous fat filled macrophages Perivascular mononuclear cells are also to be noted An *arrow* indicates the approximate border between cerebral cortex and white matter X 85 (The changes illustrated in *B* and *C* were widespread in the white matter)

leukoencephalitis are the selfsame disorder, but with a wide spectrum of pathological change—at one end of the scale the case of subacute inclusion encephalitis of Dawson (Case I, Table III) in which the cerebral cortex was affected and the white matter practically spared, and at the other end, those cases of subacute scleros-

the acute and subacute forms *are qualitatively the sa*
the key to the differing degree of involvement of the
in the two *forms may be a differing degree of dam*
dendroglia by virus. Two of us (M. G. S. and L. v. B.
the relative ease with which herpes simplex has been i
the acute disease in contrast to the failure to isolate
the subacute, although active, disease constitutes stro
that the subacute form is not due to the herpes sir
One of us (M. G. S.) feels that the differences in th
(see pages 190 and 191) also serve to distinguish th
subacute forms nosologically. None of us feels that *t*
form could be due to more than one virus.

Little has been written on pathogenesis of the acu
acute forms. Krücke[2] has postulated that sooner or la
causative virus reaches the central nervous system,
way of nerves or the blood stream, it gains the ce
fluid, then re-enters the central nervous system, as ma
(1) necrosis of superficial laminae of the cerebral
continuity of perivenous inflammatory-cell *collections*
inges with those in the cortex and white matter, a
cases of longer duration, the presence of ependymitis
observation of areas of recent inflammatory reaction
transudates of plasmatic fluid—next *to areas in the pro*
ing, Krücke concluded that repair is continually und
that in contiguous areas fresh changes frequently brea
In some of our more longstanding acute cases, we, to
cent necrosis in some areas and advanced reaction in o

That the virus is spread by the cerebrospinal flu
reasonable assumption, for in rabbits which receive
plex virus *inoculation in the cornea,* with the virus tra
sumably by way *of the trigeminal nerve, the sites of p*
of the encephalitis are often in regions which may rea
reached through the cerebrospinal fluid, namely the hi
formation, basal parts *of the cerebrum,* occipital lob
brain.[a] From the frequent tendency in the acute ca
pathological process to occur predominantly or exclusi
side of the brain—in 50 per cent of our cases as judge

autopsy changes—one would assume the existence of a local portal of entry other than, or in addition to, hematogenous spread Because of the striking predilection of involvement of the limbic lobe, one might be tempted to assume that the olfactory tract is a portal of entry, with the virus traveling in tissue spaces, perhaps chiefly perivascularly, but we have no direct evidence in the support of such an assumption Although the olfactory tracts in all 5 cases in which they were examined showed inflammatory reaction, it was minor and the sampling too small to allow an analysis of the olfactory system as a possible portal of entry Sampling of the spinal peripheral nervous system in our cases was also meager Radiculoganglionitis was observed in one instance (Fig 20) In one of his cases (26 days' duration) Krucke[illegible] noted severe radiculoganglionitis combined with myelitis of a necrotic nature, mainly perivenous in distribution, with the only other pathological change consisting of necrosis of the hippocampal gyrus Futhermore, Brain, Greenfield and Russell[illegible] have been struck in their Case 2 by the abundance of inclusions in the cervical enlargement of the spinal cord Such observations suggest the possibility of spread by way of the peripheral nervous system Krucke further commented that low grade peripheral neuritis was frequent in the subacute cases he studied As brought out in the foregoing text, others, too, have observed radiculoganglionitis or peripheral neuritis in subacute cases,[illegible] and in one instance radiculoganglionitis dominated the clinical picture[illegible] Radiculoganglionitis has been observed in about one third of the cases of subacute sclerosing leukoencephalitis (L v B)

The extraordinarily severe involvement of the cerebral white matter in some of the acute and subacute cases raises the question as to its cause It seems to us likely that in the acute cases the changes may be due to direct invasion by the virus The presence of inclusions in the nuclei of oligodendroglia would support this opinion Direct invasion of the white matter by a virus might also be the basis of white-matter involvement in some cases of the subacute form Duration of illness may be an additional factor Whether or not the white-matter involvement may also represent an isoimmunization process would be difficult to establish

9. SUMMARY AND CONCLUSIONS

There are several viral diseases in which Type A nuclear inclusions constitute a conspicuous feature, which, taken in context, allow a presumptive diagnosis to be made. The inclusions themselves are not diagnostic for they are of the selfsame appearance in different diseases. An exception is salivary-gland virus disease in which the nuclear inclusions are huge and are often accompanied by cytoplasmic inclusions. Giant-cell pneumonia is also an easily diagnosed entity because of the multiple inclusions in the nucleus and cytoplasm of giant cells

In chickenpox—herpes zoster the point to be emphasized is that inclusions may occur not only in the epidermis, but also in cells of the submucosal and myenteric plexuses, neurilemmal cells of nerve twigs, capsular cells in dorsal-root and sympathetic ganglia, and in nerve cells of dorsal-root ganglia. In chickenpox, inclusions may also occur in the lung and numerous other viscera The nosological position of Goodpasture's pneumonia is not known, but it may represent a visceral form of measles

B virus infection is a rarity contracted through exposure of abraded skin to virus-laden monkey's saliva. The virus is pantropic, and in the central nervous system the most pronounced lesion consists of severe transverse myelitis. Nuclear inclusions have been found only in animals inoculated with human material obtained post mortem.

Acute and subacute encephalitis characterized by the presence of intranuclear inclusions formed the major groups we analyzed The large number of cases available allowed nosological reappraisal

Herpes simplex infection acquired at the time of birth affects predominantly the viscera, especially the liver and adrenal cortex, and produces only minor changes in the central nervous system In infants 8 days of age or older as well as in adolescents and adults, the central nervous system almost always bears the brunt of the viral attack The pathological picture in the acute form is so distinctive that it allows the prediction of the presence of nuclear inclusions of the herpes simplex type before they are found

Subacute inclusion encephalitis and subacute sclerosing leuko-encephalitis are considered the same disorder, but at different ends of the spectrum, the cerebral cortex being mostly involved in the inclusion form, and the white matter in the sclerosing leukoencephalitic form

There was lack of unanimity among the authors of this paper as to whether the subacute form may represent smoldering herpes simplex infection. The pros and cons are presented in the text.

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DISCUSSION

Dr Druckman, Houston, Texas: Do you believe that the inclusion bodies are encephalitis virus particles or something else? I would like to ask the same question regarding inclusion bodies that have been found in cases of parkinsonism

Dr. Haymaker. In answer to the first question, the evidence based on electron microscopy of herpes simplex-virus-infected tissue cultures indicates that the homogeneous form of inclusion contains elementary bodies, and that the granular form contains few, if any As I understand it, the granular inclusion is composed wholly of altered nuclear chromatin, and the homogeneous inclusion, partly so. As to the inclusions in the parkinsonian state, they represent a slowly progressive hyaline degenerative change in the cytoplasm As the material accumulates it becomes laminated The outer laminae are strongly refractile, and the central core still more so For unknown reasons the outer laminae are alkaline, and the central core acidic By Heidenhain's azan method the central core stains red, the pericentral laminae, purple or blue, and the peripheral lamina, faint blue These structures, which measure 5 to 25 microns in diameter, are often called "Lewy inclusions" There is no evidence that they contain virus A good article on the subject is that by Greenfield and Bosanquet (*J Neurol, Neurosurg & Psychiat*, *16* 213, 1953)

Dr Casals, New York I would like to ask Dr Haymaker whether he has any data as to whether herpes simplex encephalitis is a primary infection or a recurrence Are there any antibodies

present at the start of infection, or do they develop afterward?

Dr. Haymaker: I might say that in our 53 cases, of which 9 were proven, in not a single instance were herpetic vesicles seen in the skin or elsewhere. This might be an indication that no immunity against the herpes simplex virus existed. Extensive surveys carried out in this country and Sweden* have shown antibodies in the blood stream in about 95 per cent of individuals. We do not have data on antibody studies in our series, and hence do not know whether the infections were primary or secondary. Dr. Lennette could answer that question better than I.

Dr. Lennette: I don't know how much of an answer I can provide. So far as I know, all instances of a herpetic encephalitis are generally considered to be primary infections. Buddingh's recent work in New Orleans has shown that antibodies to herpes simplex virus are present in increasingly larger proportions of the population with increasing age so that by the age of 15 years or so about 90 per cent of the population possesses antibodies to the virus. Our own serologic surveys are in essential agreement with Dr. Buddingh's findings.

I think that the cases reported in the earlier literature were uncovered primarily by chance, i.e., CNS material which happened to be available to a virologist was inoculated into laboratory animals and the virus was recovered. It was only later that it was recognized that the occurrence of fatal encephalitis in these younger people apparently represented primary infection with the virus. The occurrence of CNS symptoms due to herpes simplex virus infection in older individuals may also be presumed to be a consequence of primary infection, i.e., the disease probably occurs in the infrequent individual without prior infection with the virus and hence without antibody. I know of one physician whose wife developed a meningoencephalitis; the herpes simplex virus was recovered from her spinal fluid early after the onset of the illness and it was also demonstrated that antibody appeared and increased in titer during the course of her illness.

Dr. Rubin, Houston: In how many cases in which death occurs from a variety of causes could herpes simplex virus be isolated

from the brain?

Dr. Haymaker: I have no experience in that field and will turn the question over to someone else Dr Robbins, will you please comment?

Dr. Robbins: I don't know that I can add very much to what has been said I have heard that there is at least one instance of a patient killed in an automobile accident, previously healthy as far as anyone knew, from whose brain herpes simplex virus was isolated There were a number of experiences, as Dr Haymaker mentioned, in the days when they thought that Von Economo's encephalitis was due to herpes simplex infection—when herpes virus was isolated from human spinal fluids We no longer believe that this was related to their disease. However, I must say that one thing has intrigued me There have been a great many, possibily several hundred, spinal fluids tested in tissue culture under circumstances in which we should have been able to culture herpes virus. In our own laboratory we have done about 125 and not a single herpes virus has been isolated nor do I know of any one else who has isolated this virus from cases of aseptic meningitis Therefore, I don't think you will find it in the spinal fluid frequently It might be present in tissues, however

Dr. Rubin: Does one get rises in titer?

Dr. Robbins There is a certain number of aseptic meningitides in which a rise in herpes antibody does occur, but I don't know in what percentage Dr Lennette may have more data on that.

Dr Lennette We find such instances unusual, probably less than one percent

Dr Robbins: Unfortunately, our series has not been screened for herpes by serologic methods The point that was raised whether encephalitis is a primary or secondary infection is a most interesting one When we tried to get data concerning this we found very little in the literature Most of the cases didn't have adequate serologic studies and this is what one needs in order to settle the issue Even if only a very small percentage of the population has no antibodies in adult life, this still would be enough to account

for all of the reported cases, and more, of herpes encephalitis I have always assumed that it was primary, but it might be more analagous to zoster. Zoster is probably a recurrent form of chickenpox infection and with it central nervous system involvement often occurs, in contrast to the primary infection where it is rather unusual.

CURRENT TRENDS IN THE CONTROL OF NEUROTROPIC VIRAL DISEASES

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IN discussing the problem of current trends in the control of neurotropic viral diseases, I am going to limit my remarks to work that has been reported and is still in progress in our attempts better to control the incidence of two of these diseases namely, rabies and poliomyelitis

RABIES

Vaccination against rabies following exposure is practiced by public health authorities throughout the world, and is well recognized to have lowered the mortality rate The phenol-inactivated fixed virus vaccine first proposed by Semple[1] in 1911 is probably the one most widely used in the United States today However, although properly prepared rabies vaccines have saved many lives, they suffer from two major disadvantages they do not act fast enough to be of value in cases where severe, deep wounds have been inflicted by a rabid animal, and, second, their use involves the risk of neuroparalytic accidents

The careful studies of Otten[2] showed that there was little advantage in treating cases with anti-rabic vaccine when the exposure to rabies was so severe that the incubation period was less than thirty days These observations were supported by the findings of Ghodssi[3] and Baltazard[4] in the treatment of wolf bites in Iran

Babès and Lepp[5] were the first to use serum prophylaxis as an

adjunct to anti-rabic vaccination. This approach is based on the theory that antiserum provides quick protection in the period before the vaccine becomes effective. Other pioneers in this work were Fermi[6,7] in Italy; Shortt *et al*,[8] and Covell *et al*,[9] in India; and Proca and Babès[10] in Rumania.

More recently Hoyt,[11,12] Mabel,[13] and Koprowski and his associates[14,15] in our laboratory, have shown conclusively that antirabies serum has definite protective capacity in the treatment of exposed laboratory animals. The studies of Koprowski, Van der Scheer and Black[15] are particularly important, because they paralleled as closely as possible the natural course of exposure and treatment, and were sufficiently extensive to give significant results Hamsters and guinea pigs were exposed by the intramuscular injection of street virus. The antiserum was obtained from either rabbits or sheep hyperimmunized with chick embryo adapted or rabbit brain fixed viruses. A single inoculation of antiserum was found to give effective protection to animals infected twenty-four hours previously. In contrast, a course of fourteen injections of vaccine started twenty-four hours after infection regularly failed to protect. Combined antiserum and vaccine therapy gave essentially the same results as serum alone If antiserum was administered later than seventy-two hours after virus exposure, little or no protective effect was found.

Sellers[16] in 1953 reported on four years of experience with antirabies serum in heavily exposed individuals. Sixty-eight persons were treated with antiserum, followed in each instance by a course of vaccine. No deaths from rabies occurred, and except for scattered mild serum reactions, no side effects were noted Later, Starr[17] announced that hyperimmune serum for human prophylaxis had received sufficient trials in Georgia to warrant its use in those cases where severe lacerations and deep wounds were inflicted about the face and hands Some seventy four persons were treated with hyperimmune serum coupled with vaccination, and no deaths were recorded Starr states that, assuming the natural susceptibility of man, approximately ten patients might have died from rabies had they not received serum.

Results of evaluations of rabies antiserum in non exposed

human beings have been published by the Expert Committee on Rabies of the World Health Organization.[14] Following a single dose of hyperimmune serum, antibodies appeared in the blood within one day, persisted at a good level for at least ten days, but dropped slightly by the fourteenth day, and were present in most persons on the twenty-first day. There was no indication that serum or vaccine, when both were used, interfered with each other. Only those individuals receiving hyperimmune serum followed by twelve daily injections of phenolized vaccine had early and persistent antibody titers throughout the twenty-eight day observation period.

Conclusive and dramatic proof of the value of combined serum therapy and vaccination has come from Iran. Baltazard and his associates[17] reported results which may be summarized briefly as follows:

During the night of August 22, 1954, a large rabid wolf invaded the village of Sahane, Iran, where it bit twenty-nine persons within a few hours. Twenty-seven of them were taken at once by truck to the Pasteur Institute at Teheran, where they were treated, some twenty-eight and some thirty-two hours after being bitten. The other two men arrived later: one was treated as late as 100 hours after the bites.

Alternate persons bitten *only* on the limbs and trunk were given serum plus vaccine or vaccine alone. They all survived.

Eighteen individuals suffered severe head wounds. Of these, 5—(Series A)—were given two injections of hyperimmune serum on the first and fifth days, and phenolized vaccine for 21 consecutive days thereafter. All of these survived. 7—(Series B)—received a single injection of serum and a 21-day course of vaccine. Only one of these died. 5—(Series C)—received only the 21-day course of vaccine, and three died of rabies. An exceptionally severely bitten case—a six-year-old boy who had a crushed parietal bone and torn dura mater—was given six injections of serum at forty-eight hour intervals and vaccine for twenty-one days and survived.

The mortality rate among those receiving vaccine alone was the same as had been observed at the Institute Pasteur, Teheran.

during the previous fifteen years; that is, 40 per cent with bites on the head and face died.

The addition of a single serum injection to the vaccine treatment reduced the mortality rate to one in seven, while there were no deaths when two injections of serum were given The most unusual case was that of the six-year-old boy with the crushed parietal bone and torn dura mater, who survived after six injections of serum and a course of vaccine.

The vaccine used during this episode was of the type routinely prepared at the Pasteur Institute in Teheran, consisting of 5 per cent infected sheep brain tissue inactivated with 0 6 per cent phenol The rabies antiserum, prepared at Lederle Laboratories by hyperimmunizing rabbits, had been furnished by the World Health Organization for trial in such an eventuality as that described above. The dosage was about 0.65 ml per kg. of body weight given intramuscularly into the buttocks, with a maximum of 50 ml. injected at any one time.

Indications for Antiserum Treatment. The decision as to whether or not antirabies serum therapy should be instituted must be determined by the combination of events in a particular case. The Expert Committee on Rabies of the World Health Organization has reviewed the various possibilities following exposure and defined the indications for treatment[illegible] Antirabies serum should be considered in every case of severe exposure It should be administered within the shortest possible time, preferably within twenty-four hours: ¼ ml. per pound of body weight intramuscularly into the buttocks If treatment is delayed beyond twenty-four hours, and the exposure has been extreme, the serum dosage may be doubled or tripled. Since serum alone is not capable of stimulating antibody production, it should always be given in conjunction with vaccine. The chief function of antiserum is to establish protective titers of antibody as soon as possible, and thus "to bridge the gap between exposure and the time when vaccine induces an effective antibody response."[illegible] Present evidence indicates that vaccine therapy may be initiated twenty-four hours following the administration of serum with no untoward interfering effects.[illegible]

Recent Studies with Chick and Duck Embryo Rabies Vaccines. Neuroparalytic accidents during treatment with brain tissue rabies vaccines have been an ever-present hazard Recent figures quoted by Pait and Pearson[a] reveal that such accidents occur approximately once in 500 to 600 antirabic treatments In order to avoid the sensitization to brain tissue believed to be the cause of postvaccinal reactions, efforts have been made to provide vaccines essentially free of nervous tissues by growing the virus in either chick or duck embryos

Flury Virus Chick Embryo Vaccine. The Flury strain of rabies virus was isolated by Leach and Johnson[a] from the tissues of a girl who died of the disease, and was established directly in brain to-brain passages in one-day-old baby chicks Koprowski and Cox[a] adapted the strain to chick embryos, and showed that it became attenuated on serial passage Living Flury strain virus of about the fiftieth chick embryo passage is now widely used for the immunization of dogs A further loss in virulence occurred abruptly between the 176th and 182nd chick embryo passages, and virus from these passages, known as High Egg Passage (HEP) does not produce clinical signs of infection even when inoculated intracerebrally into rabbits, dogs or young adult mice However, the HEP strain has retained its ability to produce lethal infection when inoculated intracerebrally into suckling mice less than eight to ten days old. Koprowski and Black[a] have shown that the HEP Flury virus is effective and safe for the immunization of both dogs and cattle. Because of its minimal virulence for laboratory animals, the HEP virus was chosen for the experimental immunization of man.

Our laboratory is greatly indebted to Dr John P Fox and his associates at Tulane University for the many observations they have made on the use of HEP virus in man Initial experiments,[a] carried out with large intramuscular inocula (2 gm of chick embryo tissue in a 3 ml. volume), showed that both the frequency and degree of antibody response were related to the total mass of tissue inoculated These findings were interpreted to mean that HEP Flury virus, while living, did not multiply in man, and thus the antigenic stimulus was derived solely from the amount of

virus antigen actually injected. Large quantities of chick embryo tissue (12 to 20 gm.) were necessary to insure uniformity of neutralizing antibody response. With these rather discouraging results, subsequent work has been directed towards finding a better route and intervals of inoculation. There apparently is no problem concerning the safety of the virus, since to date more than 600 volunteers[7] have received primary courses of HEP Flury vaccine consisting of one or more inoculations. In two instances immediate anaphylactoid reactions were observed following large intramuscular inoculations. The Flury virus itself has not given rise to any recognizable reactions.[*] In addition, more than sixty men have received booster inocula at intervals of from five to thirty months after primary immunization without showing any signs of sensitization to the chick embryo tissue. The most recent work has indicated that the intradermal route may be superior to the intramuscular, and that three or four doses intradermally (0.04 gm. each) with a five-day interval produced a good serological response in eighteen of nineteen individuals. However, as good or even better results were obtained when the commercial Semple type vaccine was given in the conventional fourteen dose course, or by four inoculations with five-day intervals. Furthermore, it was found that four doses of Semple vaccine at five-day intervals did just as well as did fourteen daily doses of Harris vaccine. It should be remembered, however, that such vaccines carry the added risk of post-vaccinal accidents.

In the important aspect of rapidity of response, the HEP Flury gave better results than did the Harris or Semple vaccines. Thus of thirty-seven persons who received four or fourteen doses of Semple or Harris vaccine, ten (27 per cent) had demonstrable antibodies by the tenth day. Of eighty-eight who received the HEP Flury vaccine, forty-two, or 48 per cent, showed antibodies by the tenth day. These findings suggest that when the HEP Flury virus did elicit a response, it did so somewhat more rapidly than did the conventional vaccines.

Fox and his associates[*] have also recorded some observations concerning the ability of the HEP Flury virus to restimulate an antibody response in previously immunized persons. Of fifty four

individuals who had received a primary course of HEP Flury vaccine five to twenty-four months previously, all but four responded to a booster dose. A small intradermal dose of 0 04 gm was apparently as effective as 2 gm. intramuscularly. In another group of twenty persons, chiefly veterinarians whose past history of immunization was fairly reliable, eighteen responded to a booster dose of HEP Flury, including all those whose last previous treatment had been within twenty years. Of special interest was a laboratory worker, treated forty years previously, whose booster response was clear-cut.

Sufficient information is not yet available to determine whether primary immunization with HEP Flury vaccine is adequate for postexposure situations, but the experience with booster inocula indicates that it might be useful "to initiate and maintain non-emergency immunization in high risk groups such as veterinarians, postmen and dog catchers and, perhaps, even in the general child populations of areas where rabies is highly endemic."[*] Much more data are needed on the optimal spacing of booster inoculations, and it is hoped that studies now under way in our laboratory will give us a virus of increased immunogenicity

Duck Embryo Vaccines. Recently Peck, Powell and Culbertson[*] described an attenuated rabies virus cultivated in embryonated duck eggs. The antibody response in man was considered quite satisfactory, being early and present in most of the patients Later, the same authors[*] reported on the laboratory and clinical aspects of a beta-propiolactone-killed rabies vaccine prepared from duck embryos rather than rabbit brain tissue so as to eliminate the myelin component implicated in most postvaccinal reactions Suspensions of duck embryos with virus titers of approximately 10^6 LD[50] made effective vaccines when inactivated by 1 4000 concentrations of beta propiolactone These vaccines were stable and potent by N.I.H tests Twenty-one patients were given complete courses of fourteen injections of vaccine Tenderness at the site of injection developed in a majority of the patients and lasted from twenty four to seventy-two hours This was not a major problem Antibodies were produced in all vaccinated sub

jects within ten days. The authors point out that this type of vaccine should be used with caution in allergic subjects, especially if the allergy is to chicken-egg albumin.

POLIOMYELITIS

Now I would like to discuss the highly dramatized efforts to protect against poliomyelitis Since the Salk vaccine was introduced, many claims have been made for it, and it has been the subject of much controversy. Even now, after almost three years, enough is not yet known to say whether it will eventually be the solution to the problem, but information has emerged which is worth recounting.

Rutstein[n] reports that 15,128 cases of polio were listed by the United States Public Health Service in 1956 as compared to 28,816 cases in 1955 This sharp decrease coincided with the mass polio vaccination program which started in April, 1955, and during which approximately thirty million people, mostly children, were given one or more injections of vaccine. If the number of polio cases in the previous years had been relatively constant, the drop of reported cases from 1955 to 1956 might prove the efficacy of the vaccine. However, there have been great variations in the number of polio cases from year to year. Rutstein's[n] data show that in the years 1930 through 1943 and in 1915 and 1917 the incidence of polio was no greater than in 1956, and in many of these years was in fact lower. Beginning in 1918 there was an annual increase, reaching a peak in 1952 with 57,879 reported cases. As yet there is no explanation for the great differences from year to year, but they occur not only in the United States but in practically all parts of the world.

Perhaps more reliable information can be gained by comparing the totals of clinically recognized paralytic cases In 1955, there were 10,405 (to December) as compared to 6,565 for the same period in 1956, a decrease of approximately one third[n] This type of information is much better for evaluating the effect of polio vaccination, and thus far the swing appears to be in the right direction In line with this are the figures on the 1955 epidemic in Massachusetts,[n] which was caused almost exclusively by

Type 1 virus. A total of 130 cases was reported among 137,968 children who received a single dose of vaccine an attack rate of 94.5 per 100,000. Fifteen cases of polio were recorded among 22,673 children who received two or more doses of vaccine, or an attack rate of 66.4. In comparison, there were 553 cases of polio among 278,532 unvaccinated children, giving an attack rate of 198.2 per 100,000 It was estimated that the effectiveness of the vaccine for all cases of poliomyelitis was 53 per cent, and 60 per cent for paralytic cases These figures are comparable to those quoted by Francis and his associates[*] for protection against Type I poliovirus

Of great interest is Rutstein's observation that the intense polio vaccination campaign begun *after* the start of the 1956 epidemic in Chicago had no apparent effect on the course of that epidemic. If the vaccine had exerted a real effect, there should have been "a sharp drop in the number of cases" within a fairly short period of time As it was, "the upswings and downswings on the epidemic curve" were of the same shape and showed the same picture as are usually seen in this disease

Furthermore, all evidence now indicates that vaccination with Salk vaccine does not eliminate alimentary infection, and that vaccinated children are as likely to carry and shed living polio virus as are unvaccinated children[*] Possibly this is fortunate, but in any event only time will tell how effective the killed vaccines will be in protecting against paralytic poliomyelitis without benefit of further immunizing measures As Paul has stated[*] the answer will probably depend on where a person lives and "as such, what his postvaccinal exposure may be, because the degree of exposure to polioviruses differs in different places and according to different ways of life "

Everything must be done to insure that we are not postponing the occurrence of poliomyelitis from childhood until later in life when the disease is more severe, and paralysis and death are more frequent[*]

In all probability the present Salk vaccine can and will be improved by finding better methods of inactivation so as to retain antigenic capacity, and also by learning how to increase the

cells, although later two of four infants excreted TN virus that was cytopathogenic. This strain has been fed to at least 423 people of various ages with or without the protection of immune serum globulin or maternal antibody, and apparently has never produced viremia, fever, clinical signs of illness or contact infection. Quantitative studies indicate that as little as 450 PD_{50} of virus was sufficient to immunize certain individuals, although the customary dose has been a 1 ml amount (of 20 per cent mouse brain and cord suspension) containing anywhere from 10 to 10 PD_{50} of virus. Neutralizing antibodies have been found to persist for at least three and five-year periods of time.

The SM strain is a Type 1 virus that was first adapted from monkey kidney tissue cultures to the spinal cord tissue of mice and cotton rats. It was then carried for twenty-seven consecutive intraspinal passages in PRI (Princeton Rockefeller Institute) mice and for fourteen serial passages in chick embryo tissue cultures. It was next plaqued out by the Dulbecco technique on monkey kidney monolayer plates. Five alternating passages were made between monkey kidney and chick embryo tissue cultures, and it was again plaqued out three consecutive times. Finally, it was fed as a first or second passage in chick embryo tissue culture. This strain is cytopathogenic for monkey kidney epithelium, and may be assayed either in tissue culture tubes or on monolayers of monkey kidney epithelial cells (Dulbecco plates). Like the TN, Type 2, strain described above, the SM strain is nonpathogenic for monkeys inoculated intracerebrally. It does cause paralysis in some monkeys when injected intraspinally in high concentrations. Thus far, all those fed the SM strain have excreted virus, quite often in concentrations as high as 5.5 to 6.5 logs, in the first fourteen days after feeding. Furthermore, those fed have remained intestinal carriers of virus for long periods, up to 171 days. In a group of twenty-two infants—all under six months old and six of them ranging between ten and twenty-seven days old, the highest concentration of virus in the stools was found during the first sixteen days after feeding and the duration of alimentary infection ranged from fifty-five to 108 days with the average being about seventy-three days. In this con-

nection, Sabin has reported a maximum excretion period of 140 days for a person fed his partly attenuated Type 1—Mahoney KP33—strain. Thus far the SM Type 1 virus has been fed to at least 257 subjects of various ages, with or without the protection of immune serum globulin or maternal antibody, and apparently has called forth uniformly an immune response without causing any clinical illness. In early trials, Koprowski found that as little as two plaque-forming particles of the SM strain could initiate an intestinal infection in some children but not in others. Later studies with a higher passage level of the strain indicated that 1,000 to 10,000 plaque-forming particles must be fed to insure intestinal infection. Additional information is needed concerning the contagiousness of the SM strain. In one experiment the virus was transmitted from child to child in five of fifteen contacts "under the most intimate contact conditions." In another trial seven children who were fed the SM strain were placed in the same nursery with two infants, twenty-two and twenty-six days old, who received no virus. Those who had been fed excreted Type 1 virus for as long as seventy to ninety-nine days, and all developed neutralizing antibodies. The two contacts were kept in the nursery for one and a half and four months, respectively, and yet they failed to excrete virus or to develop antibodies.

Not as much information on the duration of antibodies following virus administration is available for the SM as for the TN strain, but they have been found to persist for at least twelve to fifteen months. It has also been observed that the SM strain interferes with the immunogenic activity of the TN strain when the two are fed simultaneously, so that it is now thought advisable to feed the viruses separately at about three to four week intervals.

The MEF 1 is a Type 2 strain that was first adapted to the central nervous tissues of mice, then to the CNS of suckling hamsters. During hamster passages, a variant developed that grew readily in chick embryo tissues. The cultural and growth characteristics of the strain have been well described by Cabasso *et al*. As far as I know, this is the only strain of poliovirus that has been fully adapted to grow in the chick embryo. Roca has shown that

this strain produced a mild attack of poliomyelitis in only one of thirty-five monkeys inoculated intracerebrally with a high concentration of virus. Slight cord lesions were found upon histological examination in only eight of thirty-eight monkeys inoculated intraspinally. Cynomolgus monkeys injected repeatedly with large doses by the intramuscular route showed no symptoms but developed specific neutralizing antibodies. Similarly, chimpanzees infected intramuscularly or orally were found to have antibodies although they did not become fecal carriers or show any signs of illness. The MEF 1 strain is almost completely noncytopathogenic for monkey kidney cells, but retains its ability to kill mice and hamsters inoculated intracerebrally, even after more than 150 serial passages in chick embryos. The above characteristics indicate that this strain of virus is greatly altered, and that it carries a number of biological tags or markers that serve to differentiate it from naturally occurring strains of virus. Thus far only the seventy-first chick embryo passage of this strain has been fed to 160 individuals. Twelve of them received immune serum globulin one hour after the virus was fed (0.14 ml. per pound of body weight).[illegible] The others were given virus alone. The results indicate that the MEF 1 strain of the seventy-first chick embryo passage level is so greatly modified that fairly large doses must be fed in order to induce an immune response, that is, from 200,000 to 800,000 mouse LD_{50} (5 ml. of a 10 or 20 per cent chick embryo suspension). No illness, no viremia, and no secretion of virus in the pharynx were found. Thus far, virus has been recovered from the stools of only three persons, and then in low concentrations and for periods not exceeding sixteen days. In all cases the amount of virus in the stools was so small that it was detectable in mice only after enrichment by tissue culture passage. There has been no evidence to show that virus recovered from the stools regained virulence for the monkey. Of thirty-two monkeys inoculated intracerebrally with either stool suspension, tissue culture material, or first mouse passage material, only one came down with paralysis. This animal had received undiluted tissue culture material derived from a stool specimen collected 11 days after feeding. None of the other monkeys became ill, and the histopath-

nection, Sabin has reported a maximum excretion period of 140 days for a person fed his partly attenuated Type 1—Mahoney KP33—strain.[illegible] Thus far the SM Type 1 virus has been fed to at least 257 subjects of various ages, with or without the protection of immune serum globulin or maternal antibody, and apparently has called forth uniformly an immune response without causing any clinical illness.[illegible] In early trials, Koprowski[illegible] found that as little as two plaque-forming particles of the SM strain could initiate an intestinal infection in some children but not in others. Later studies with a higher passage level of the strain indicated that 1,000 to 10,000 plaque-forming particles must be fed to insure intestinal infection.[illegible] Additional information is needed concerning the contagiousness of the SM strain. In one experiment[illegible] the virus was transmitted from child to child in five of fifteen contacts "under the most intimate contact conditions." In another trial[illegible] seven children who were fed the SM strain were placed in the same nursery with two infants, twenty-two and twenty-six days old, who received no virus. Those who had been fed excreted Type 1 virus for as long as seventy to ninety-nine days, and all developed neutralizing antibodies. The two contacts were kept in the nursery for one and a half and four months, respectively, and yet they failed to excrete virus or to develop antibodies.

Not as much information on the duration of antibodies following virus administration is available for the SM as for the TN strain, but they have been found to persist for at least twelve to fifteen months.[illegible] It has also been observed that the SM strain interferes with the immunogenic activity of the TN strain when the two are fed simultaneously, so that it is now thought advisable to feed the viruses separately at about three to four week intervals.[illegible]

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ological findings were similar to those seen in monkeys inoculated intracerebrally with the seed inoculum used for vaccine production. Virus recovered from stools retained its growth characteristics in mice and chick embryos

The Fox strain is a Type 3 virus that was isolated by Dr. John P Fox of Tulane University from a patient with nonparalytic poliomyelitis. This strain, which grows thus far only in monkey kidney tissue culture, has been "purified" by being plaqued out *six consecutive times by the Dulbecco technique. It is highly cyto*pathogenic for monkey kidney tissue culture cells. However, it is nonpathogenic for cynomolgus monkeys inoculated intracerebrally or intramuscularly. Intraspinally, in high concentrations, it will produce paralysis in an occasional monkey. Thus far the strain has been fed to only thirty-one people, so that it has not been as well studied as the other strains No ill effects have been seen. Virus has been recovered from stools for at least thirty days, but the recovered virus has not regained virulence for monkeys No viremia has been observed in the treated cases, and specific neutralizing antibodies have appeared on or about the eighth to *tenth day after feeding*

Since this paper was prepared, Dick and his associates[illegible] have described clinical trials in North Ireland in which the TN and SM strains of poliovirus were fed. There are some discrepancies between their data and the data reported on trials conducted in the United States. As yet these differences in findings have not been resolved, and the whole matter will have to be investigated thoroughly before further comment can be made.

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DISCUSSION

Dr. Fahlberg, Houston, Texas: Is it valid to use the interpretation that following mass inoculations we show an apparent decrease in the incidence of poliomyelitis? This may be true only in children but not apply to adults.

Dr. Cox: I don't have the breakdown on age groups We think the results in Massachusetts would indicate that the trend is in the right direction, because there was apparently a reduction in the total number of polio cases, roughly about a 50% drop in the paralytic type, but I think we all agree that it is still a bit too early in assuming that this is a final answer, or a valid result. There is, no doubt, I think, that the killed vaccine is showing an effect. How long the duration of immunity is remains to be seen. Does anyone else wish to make any further comments?

Dr Robbins Cleveland, Ohio: I think that there is no doubt that the vaccine will prevent paralytic polio.

Dr Kurland, Bethesda, Md.: I wish to point out that the apparent discrepancies in ratio could be due to the large increase in the birth rate—more than twenty million more children were born in the postwar years than in the comparable prewar period

Dr Fields, Houston, Texas: Has there been any evidence that the Salk vaccine has produced a viremia?

Dr Cox There were of course, instances of viremia with use of the early vaccine preparations. The present product, I think, is safe The question is—how potent is it? As far as I know there is no danger of getting viremia or poliomyelitis from the product that is being produced now, but we do recognize the fact that potency has possibly taken a sharp decline because of the present product being subjected to three consecutive filtrations Some manufacturers are having trouble obtaining good potency, not only in this country, but in Canada and England as well.

Dr Fahlberg At a local blood bank they will not draw blood

from any person who has had a polio vaccination in the previous six weeks. I am not yet certain as to whether or not this is the national regulation

Dr. Cox: I have not been aware of any such regulation (asks several discussants if they had heard of this· All spoke in the negative). I think that an important fact of which we must be aware is that people who have received the Salk vaccine can still become intestinal carriers with naturally occurring strains of virus. This may turn out to be an advantage

Dr. Blattner, Houston, Texas: Then this information points to the fact that when the person receives live virus he may become a carrier, whether he has received Salk vaccine or not

Dr. Cox: We don't have much data on that, but one thing I would like to point out is this—we at least are anxious to get our strains as modified as much as possible so as to induce antibodies *but still not excrete virus for long periods of time* Our Type 2 chick embryo virus strain seems to be the most modified that we have because the amount of virus that can be picked up from the stool is very small We have to use the procedure of tissue culture inoculation in order to get enough virus produced to bring down mice. If we inoculate stools directly into mice or into monkeys we fail to find virus We think it more ideal to have virus strains that do not produce virus in the stools for too long a period of time In people who have received the living virus there are indications that when one goes back and refeeds these same people, it is more difficult to set up an intestinal carrier state a second or third time Koprowski has done some studies in which he fed twelve people the Type 2 strain and approximately six months later went back and fed them a second time, and on this latter occasion only two of the twelve became excretors of the virus Then when these people were re-fed for the third time, some time later, only one out of twelve showed any virus excretion and this occurred for only a few days Sabin has also noted that people who have been naturally immune or people who have received living modified viruses show a much decreased incidence of the intestinal carrier state when fed a modified virus strain later on In other words, there is a good deal of information pointing to the fact that when

you receive living virus the possibility of becoming an intestinal carrier on reinfection is greatly diminished

Dr. Burdon, Houston, Texas: The preparations of attenuated virus is a long expensive process, and aren't we going to have to give more inoculations? We are looking forward to vaccines of that kind, but won't it be a long time before such vaccines are ready, and won't they be expensive?

Dr. Cox: We are not yet ready for commercial production We hope that we will be ready somewhere around 1958 to 1960 We can readily get antibodies by feeding live viruses by the oral route. In our own laboratory we would prefer to get all these strains adapted to some type of cells other than monkey tissue cells. We would like to have a biological tag on them so that we could readily differentiate our strains from wild strains It is very slow work. We have only one such tagged strain at the present time; the MEF 1, Type 2 We feel that we are making progress on Types 1 and 3 but we don't have them fully adapted and tagged as yet Once we accomplish these essentials we can readily turn out large quantities of vaccine with a relatively small amount of material One doesn't have to have the volume because the antigenic mass is not required that would be required in a killed vaccine. This is so because one depends upon the virus multiplying in the gut to a certain extent and thus producing the antigenic stimulus required. The main thing is to see to it that the virus doesn't multiply too long or regain its virulence. I think that this can be done. It is a terrific challenge and it is not an easy thing to do, but we believe it can be done. Personally, I don't believe that our present strains that we are working with represent the final answer in this respect.

Dr Alvord, Houston, Texas: Regarding post-infectious or post-vaccinal encephalitis—are there any new current views on the etiology of this? Is it an allergic reaction, or is it actual virus infection?

Dr Blattner: Dr. Cox, do you want to answer that question?

Dr Cox No, I'll turn that over to someone else. Perhaps Dr Robbins would like to answer Dr. Alvord.

Dr Robbins: I think you are all familiar with the various

theories. There is a possibility that another virus is the cause. Still another possibility is that it is due to auto-allergy to brain tissue There are viruses that produce a demyelinating process similar to that seen in post-infection encephalitides There is no conclusive proof at the moment of the etiology of these disorders except in mumps where the virus has repeatedly been isolated from the spinal fluid.

Dr. Blattner: Dr. Haymaker, do you care to comment?

Dr. Haymaker. Dr. Robbins handled that question so well that I haven't a thing to contribute.